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ZOOLOGICA

SCIENTIFIC CONTRIBUTIONS
OF THE
NEW YORK ZOOLOGICAL SOCIETY

AUG 1957
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VOLUME XXI

Part 4

Numbers 19-23

PUBLISHED BY THE SOCIETY
THE ZOOLOGICAL PARK, NEW YORK

December 31, 1936

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19.

The Southwestern Desert Tortoise, *Gopherus agassizii.*

CHAPMAN GRANT.

Several enterprising youths in the vicinity of Hodge, San Bernardino County, California, have started commercializing the desert tortoise, *Gopherus agassizii.* May 8-11, 1935, was spent examining several small collections and Robert Heckly's two hundred specimens which were for sale to tourists. On another trip, October 2-9 inclusive, one hundred more specimens were examined. Most were in captivity, some were captured and some shells of dead specimens were studied. The favorable hours during which the tortoise ventures abroad were devoted to field work. Some observations that have not been seen in print are offered, together with additional data which may prove of interest.

ASYMMETRY.

Examination of plastrons showed that the suture between gulars is offset to the right from one to five millimeters, making the left gular wider. It does not protrude farther, but has a wider base and grows farther back on the plastron. Among 366 specimens, 331, or 90%, had the suture on the right, 23, or 6%, were median and 11 females and 1 male, or 3%, had a left suture. The 23 with central sutures were about evenly divided between the sexes. Plate XXIV of Ditmars' "The Reptile Book" (1907) illustrates the asymmetry. Additional evidence is shown in 15 plates by O. P. Hay in "The Fossil Turtles of North America." The left gular is larger in 7 plates and in no case is the right larger. Asymmetry of the gular scutes is in contrast to the median suture of the symmetrical underlying epiplastral bones. The gulars are either forked or worn chisel-shaped and recurved to a varying degree without apparent reference to age.

DIMORPHISM.

In the males secondary sexual dimorphism consists of larger size, longer gulars and tail, smaller pelvic clearance measured from seam of anals to edge of rear marginals, thickened anals and concave plastron, characters which make their appearance as the juveniles exceed 12 or 14 cm., with the difference in tail length showing first. The sexes have similar proportions except that the male exceeds by 2% in length of gulars and 5% in tail length, with a 2% smaller pelvic clearance. Among 366 specimens 19 females, 7 males and 4 young had major scute atavisms or abnormalities. One female was nearly circular, having narrow costals; another was elongated, having extra scutes, and a third was exceptionally high. The young are light, but color apparently has no sexual significance. Two partial albinos were seen with carapace, legs and toenails olive gray. The legs of

one large male were orange and black instead of the usual gray and black. Varying recurvature of the marginals over the hind legs and the amount of curving-under of the pygal as well as the considerable variation in number and arrangement of the inguinals are without apparent sexual significance. The color of the iris is brassy or brown or these two colors mottled, with about 80% brassy, 10% mottled and 10% brown without correlation to sex or age.

Dimensions.

Table of dimensions in cm.

	30 adult females	Largest female	30 adult males	Largest male
Carapace length....	24.27	28.6	28.3	33.7
Carapace depth....	10.4	11.5	12.	14.6
Greatest width.....	19.8	22.9	23.17	26.7
Plastral concavity..	....	....	1.47	1.8
Gular length.....	4.17	5.3	5.47	7.2
Pelvic clearance....	3.6	4.3	3.9	4.5
Tail length.....	2.27	2.8	4.00	5.8
Weight in gms.....	....	2,765.	....	5,809.

Description.

Occasionally an old female is found with the caudal worn flat or even cut through, leaving a semicircular nick, as a result of being rubbed by the anal plates of the male. Growth rings are usually visible on full grown specimens, but disappear with age. Scutes become concave by thickening along their edges when age is attained. Occasionally spines about as long as the toenails, extending around the heel, give a starlike contour to the hind foot. The retracted thighs are protected by pointed tubercles, usually in the form of a circle around a large central spine.

Habits.

One old male was seen with tail hanging down and slowly swinging from side to side with each elephantine step. A large male pushed himself along the ground carrying Robert Heckly and one persisted in crawling under the rung of my chair and tilting me. The little Heckly girls staged "roller skating" races, each standing on two tortoises.

Males utter grunting sounds while courting and recognize one another immediately, possibly by scent which may come from glands beneath the bulbs of the jaws. The long gulars are used by fighting males, which charge one another, their heads retracted. When one manages to ram his protruding gulars into the groin or anterior opening of the opponent's shell, he lifts and twists, often upsetting his antagonist. An overturned tortoise flaps one front leg violently, stopping it suddenly at the most forward position, the momentum jolting it slowly around so that any fixed object within reach may be utilized to right itself. Males may perish if they cannot turn over, since the hot sun is soon fatal. When asleep they sprawl legs and necks in the most grotesque positions, appearing dead. They become tame and inquisitive and seem to enjoy having their heads rubbed.

Fantastic stories of homing instincts may contain a grain of truth as the tortoises usually return to the same burrow nightly. Accounts of treks

or migrations and turtle towns are prevalent. I once saw what appeared to be a migration of *G. berlandieri* in southern Texas and have noted that tortoises are numerous only in restricted localities.

Robert Heckly furnished the following data: "In June, 1934, each of three females laid five eggs and one laid two in holes dug by the hind feet. It took an hour to dig the hole and fifteen minutes to lay the eggs. After depositing an egg, a foot was thrust into the hole, seemingly to roll the eggs from side to side or to pack them in. The hole was then filled and covered by the same process. Four from one female hatched in November, the fifth hatched the following March. The rest were infertile. By October the adults feed infrequently and by December have stopped entirely. They are placed in the cellar under canvas until March when they are brought out during the heat of the day to begin eating and mating, which is over by the last of April. They lay the latter part of June. They have been known to eat dried jackrabbit meat."

Tortoises are eaten during lean years, but no one seemed enthusiastic about the meat. I found the shells of three near the ashes of a small fire in the desert with evidence that the largest carapace had been used for a cooking vessel—an Indian custom. Escaped or liberated specimens are found with a hole drilled in the edge of the shell or a portion of the edge broken out.

Mr. Thomas Hallinan wrote an interesting article on *G. polyphemus* of Florida. Contrasts between the two species are that the present species makes no effort to escape by entering the burrow even if at the entrance, whereas the Florida species scrambles to escape. The burrow is much less steep, usually 12 degrees, starting against a small bank of wind-swept sand at the base of a bush, and humidity is disliked. In Florida the burrows are in damp ground and average fourteen feet long. In California the four-foot burrows are much straighter, the occupant frequently being visible. Never more than one tortoise and no associates, comensals or parasites were found in any burrow in California, whereas the Florida species has associates and parasites. Captive specimens at Thermal are subject to very active ticks which attack the sutures of the carapace instead of the skin and do not attach themselves.

Water Storage.

Persons in the desert say that the tortoise stores water in sacs in each side of the body. The earliest reference noted is by E. T. Cox in the *American Naturalist* of 1881. He stated that while preparing a museum specimen he found on each side a large membranous sac filled with clear water. Dr. R. D. Harwood of San Diego State College kindly dissected two specimens for verification. He found the body cavity full of coelomic fluid and a large, simple, urinary bladder of delicate membrane lying ventrally across the plastron. The sides of the bladder, tucked up between the intestines and marginals on each side, have apparently been mistaken for independent sacs. Dr. Miller thinks that water is produced by metabolism.

Young.

A captive tortoise laid six eggs at Victorville on June 9, 1935. On October 6, I purchased three of these eggs: one pipped, one nearly ready to hatch and one infertile. The remaining eggs were not for sale, but were sufficiently uncovered to see that they were fertile. The three eggs weighed and measured as follows: pipped egg 27.2 gm., 42 x 36.5 mm; fertile egg 30.2 gm., 42 x 37.5 x 35.7 mm; infertile egg with a large air bubble, 27.7 gm., 42 x 36 mm. The shell is hard and unyielding and when blown weighs

3.97 gm. Unfortunately the fertile egg was broken but the embryo was preserved. It was not ready to hatch, having blood vessels surrounding the white and the large yolk. Both this and the living specimen show great scute abnormality. No evidence was found that the eggs are buried singly as stated by Dr. Miller. The embryo lies partly curled and at right angles to the long axis of the egg. It has an egg tooth at the center of the snout with which it makes a ragged opening, parallel to the long axis. The egg tooth apparently is not shed, but flattens out after several months. The head is presented at 45 degrees, the eyes closed, but it opens its mouth threateningly if touched. The hatchling is wider than long, but unrolls and appears nearly normal in five days. A dozen captive babies were observed, all showing pugnacity, bucking forward with open jaws and hissing if touched on the carapace.

The baby hatched October 7, bearing a yolk sac and a small mass of clear, thick, jellylike substance under the plastron. The jelly dried up and the yolk was absorbed in two days. The baby measured in mm:

	1935			1936	
	Oct. 7	Oct. 8	Oct. 11	June 18	Aug. 15
Length	36	41	44	44.5	48
Width	39	37.5	36.5	36.5	42.5
Weight	..	..	19.7	..	..

It drank frequently, but ate almost nothing until May. Efforts to feed were clumsy at first, but by August it was eating tender vegetables, bananas, bread, dry grass with a generous proportion of dirt and slacked lime. It started to grow rapidly in July.

Mrs. Pajanew at Cinco had four that hatched on September 2. One had been sold, but on October 4 the remaining three weighed 23, 23.5 and 25.5 gm. Mr. G. W. Hilton near Coachella had one that hatched on September 15 and his son, John, of Thermal, reported numerous hatchings about August 15. Young specimens are soft, but when 10 cm. in length, or about five years old, seem able to resist carnivores, as several were seen with tooth marks on their shells.

Range.

The range was plotted on a map, using records from scientific books and articles, from the writer's observations and from verbal reports where three independent observers of apparent sincerity gave the same locality.

The tortoise does not occur in the Coachella-Thermal district where the temperature is probably higher than in their habitat, but Mr. Hilton's tortoises breed and reproduce well, showing that range restrictions are not due to temperature or low elevation.

The range was found to extend northward into the Panamint Valley and the Shoshone area in Inyo County. There are no records west of the Coachella Valley, so it does not approach San Diego County. It is reliably reported from the Beatty-Bunkerville line southward in Nevada and in Washington County, Utah, and the Phoenix-Florence-Tucson area of Arizona. There are a few Sonora records and one from Tiburon Island. This is a vast area, but great stretches are uninhabited.

In determining the range, tracks, burrows, droppings and dead shells are more convincing proof than a live animal encountered in an unexpected place, as there is always the likelihood that it may have escaped. At least 600 specimens were collected within two miles of Hodge in 1934-1935 ac-

cording to the census of the boys selling them. By driving to likely places, making a hurried reconnaissance and hunting only where tracks abounded the writer was able to collect ten specimens in about six hours—some in burrows and some feeding. They emerge in the forenoon and afternoon during spring and fall.

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20.

Plankton of the Bermuda Oceanographic Expeditions. VII. Siphonophora Taken During the Year 1931[1].

CAPTAIN A. K. TOTTON, M. C.
British Museum (Natural History).

INTRODUCTION.

This is one of a number of papers dealing with the planktonic contents of nets drawn at definite levels and in a definite area off the south coast of Bermuda on the Bermuda Oceanographic Expeditions of the New York Zoological Society. The area chosen was a circle eight miles in diameter, with its center located at 32° 12′ N. Lat., 64° 36′ W. Long., a point nine miles southeast of Nonsuch Island, Bermuda.[2] The depth at this locality is 1,000 to more than 1,400 fathoms. Further details concerning the nets, position, etc., will be found in ZOOLOGICA, Volume XIII, Numbers 1, 2 and 3.

Captain Totton has kindly identified the hundred-odd vials of siphonophores which I sent him, and Dr. Henry B. Bigelow and Dr. Mary Sears have furnished the faunistic notes. My own share has been only the collecting, and the gathering of the scanty field notes.

WILLIAM BEEBE.

Suborder Calycophorae.
Family Prayidae.
Subfamily Amphicaryoninae.
1. *Amphicaryon acaule* Chun 1888.

MATERIAL: No. 3174; Net 988; 1,000 fathoms; June 2.

COLOR: Lemon yellow in life.

DISTRIBUTION: This species is widespread in the tropical and subtropical belts of the great oceans (for localities see Totton, 1932, p. 330). It was first described by Chun (1888) from the Canaries, but it was not taken again in that general region until 1910, when the *Thor* took it in the Bay of Cadiz (Bigelow and Sears, in press), and in 1913, when the *Hirondelle II* took it in 38° 58′ N. Lat., 44° 55′ W. Long. (Leloup, 1933). There is only one record of it in the Mediterranean, made by the *Thor*.

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[1] Contribution No. 514, Department of Tropical Research, New York Zoological Society.
[2] For diagram of trawling area, see *Zoologica*, Vol. XXI, No. 9, page 97.

Subfamily Nectopyraminae.

2. *Nectopyramis* sp. nov?

MATERIAL: No. 31942; Net 1120; 400 fathoms; Aug. 3.
No. 311087; Net 1146; 600 fathoms; Aug. 7.
No. 311652; Net 1239; 900 fathoms; Aug. 29.
No. 311767; Net 1259; 1,000 fathoms; Sept. 3.

Subfamily Prayinae.

3. ?*Praya dubia* Quoy and Gaimard 1834.

MATERIAL: No. 31509; Net 1063; 300 fathoms; July 8.
No. 311617; Net 1226; 300 fathoms; Aug. 27.

COLOR: Transparent and colorless in life.

DISTRIBUTION: Previous records of *P. dubia* are from Australian waters, from the eastern tropical Pacific (Bigelow, 1911; 1931), and off Valparaiso (Moser, 1925).

4. ?*Rosacea cymbiformis* Delle Chiaje 1842.

MATERIAL: No. 31540; Net 1064; 100 fathoms; July 9.

DISTRIBUTION: *Rosacea cymbiformis* has been recorded from all three oceans—in the eastern Atlantic from the tropics northward to the Bay of Biscay (Chun, 1888; Haeckel, 1888; Leloup, 1933), in the South Atlantic (Leloup, 1934; Hardy and Gunther, 1935), in the Philippines (Bigelow, 1919), and in the Indian Ocean (Huxley, 1859; Browne, 1926). There are also frequent records from the Mediterranean whence it was first described.

Family Hippopodiidae.

5. *Hippopodius hippopus* Forskål 1775.

MATERIAL: No. 31305; Net 1036; 200 fathoms; June 25.
No. 31445; Net 1050; 25 fathoms; July 6.
No. 31480; Net 1055; 50 fathoms; July 7.
No. 31733; Net 1089; 50 fathoms; July 18.
No. 311252; Net 1173; 400 fathoms; Aug. 14.
No. 311301; Net 1185; 900 fathoms; Aug. 15.
No. 311473; Net 1201; 800 fathoms; Aug. 19.
No. 311499; Net 1210; 1,000 fathoms; Aug. 20.
No. 311534; Net 1214; 1,000 fathoms; Aug. 21.
No. 311618; Net 1227; 400 fathoms; Aug. 27.
No. 311963; Net 1293; 800 fathoms; Sept. 12.
No. 312066; Net 1308; 100 fathoms; Sept. 16.

COLOR: Transparent and colorless.

DISTRIBUTION: This is one of the more common species found in all the warmer oceans (for summary, see Moser, 1925).

6. *Vogtia glabra* Bigelow 1918.

MATERIAL: No. 311199; Net 1169; 700 fathoms; Aug. 12.

DISTRIBUTION: This species was originally described from the Straits of Florida (Bigelow, 1918) and has since been taken in the eastern side of the temperate Atlantic between the Azores, Canaries and the coast of Portugal and the Gulf of Gascony, as well as in the Mediterranean (Leloup, 1933; Bigelow and Sears, in press). It has never been reported from other oceans, as have other members of this bathypelagic genus.

Family Diphyidae.

Subfamily Abylinae.

7. *Abyla dentata* Bigelow 1918.

MATERIAL: No. 311652; Net 1239; 900 fathoms; Aug. 29.
No. 311767; Net 1259; 1,000 fathoms; Sept. 3.

DISTRIBUTION: This species was first described from the western Atlantic between Bermuda and the American coast (Bigelow, 1918). It has only been noted since then near the Cape Verdes (Moser, 1925, as "*A. quadrata*").

8. *Abylopsis eschscholtzii* Huxley 1859.

MATERIAL: No. 311040; Net 1133; 700 fathoms; Aug. 5.
No. 311534; Net 1214; 1,000 fathoms; Aug. 21.

COLOR: Dull crimson in life.

DISTRIBUTION: This species is widespread over the tropical Pacific and Malayan region (Bedot, 1896; Lens and Van Riemsdijk, 1908; Bigelow, 1911, 1931; Totton, 1932), the Indian Ocean (Browne, 1926), in the South Atlantic (Moser, 1925; Leloup, 1934), also in the tropical Atlantic (Chun, 1888; Mayer, 1900; Leloup, 1934). It has also been taken in the Mediterranean (Leloup, 1933).

9. *Abylopsis tetragona* Otto 1823.

MATERIAL: No. 31883; Net 1107; 400 fathoms; July 27.

DISTRIBUTION: This is one of the commoner species of siphonophores and is found throughout the warmer zones of all oceans (see Moser, 1925).

Subfamily Ceratocymbinae.

10. *Ceratocymba sagittata* Quoy and Gaimard 1827.

MATERIAL: No. 31517; Net 1062; 300 fathoms; July 8.
No. 31920; Net 1117; 1,000 fathoms; July 29.
No. 311250; Net 1174; 500 fathoms; Aug. 14.
No. 311724; Net 1248; 600 fathoms; Sept. 1.
No. 312031; Net 1305; 500 fathoms; Sept. 15.
No. 312067; Net 1308; 100 fathoms; Sept. 16.
No. 312071; Net 1308; 100 fathoms; Sept. 16.
No. 312079; Net 1309; 100 fathoms; Sept. 16.
No. 312169; Net 1322; 300 fathoms; Sept. 18.
No. 312171; Net 1322; 300 fathoms; Sept. 18.
No. 312182; Net 1330; 1,000 fathoms; Sept. 19.

COLOR: Transparent anteriorly; milky white posteriorly.

DISTRIBUTION: This species occurs in the warm parts of the North and South Atlantic (Moser, 1925; Leloup, 1933; 1934), the eastern tropical Pacific (Bigelow, 1911), the Indian Ocean (Browne, 1926), and the Malayan region (Lens and Van Riemsdijk, 1908), and in the Mediterranean, where it was taken for the first time by the *Thor* (Bigelow and Sears, in press).

Subfamily Diphyinae.

11. *Diphyes dispar* Chamisso and Eysenhardt 1821.

MATERIAL: No. 31258; Net 1020; 14 fathoms; June 15.
No. 31304; Net 1035; 100 fathoms; June 25.
No. 31356; Net 1037; 300 fathoms; June 25.
No. 31364; Net 1040; 25 fathoms; June 26.

No. 31409; Net 1045; 25 fathoms; June 27.
No. 31435; Net 1050; 25 fathoms; July 6.
No. 31481; Net 1055; 50 fathoms; July 7.
No. 31507; Net 1055; 50 fathoms; July 7.
No. 31484; Net 1056; 50 fathoms; July 7.
No. 31517; Net 1069; 50 fathoms; July 10.
No. 31598; Net 1069; 50 fathoms; July 10.
No. 31621; Net 1075; 50 fathoms; July 11.
No. 31643; Net 1075; 50 fathoms; July 11.
No. 31650; Net 1079; 50 fathoms; July 14.
No. 31732; Net 1089; 50 fathoms; July 18.
No. 31787; Net 1099; 900 fathoms; July 24.
No. 311211; Net 1169; 700 fathoms; Aug. 12.
No. 311496; Net 1206; 800 fathoms; Aug. 20.
No. 311571; Net 1218; 700 fathoms; Aug. 24.
No. 311625; Net 1230; 900 fathoms; Aug. 27.
No. 312005; Net 1285; 800 fathoms; Sept. 10.
No. 312030; Net 1301; 50 fathoms; Sept. 15.

COLOR: All transparent and colorless except No. 31650, which is described as "siphonophore with long yellow chain."

DISTRIBUTION: This species has been recorded from the warm zones of all oceans.

12. *Chelophyes appendiculata* Eschscholtz 1829.

MATERIAL: No. 311122; Net 1154; 700 fathoms; Aug. 8.
No. 312071; Net 1308; 100 fathoms; Sept. 16.

DISTRIBUTION: This is the commonest of all siphonophores, and is found in all oceans.

13. *Chelophyes contorta* Lens and Van Riemsdijk 1908.

MATERIAL: No. 311534; Net 1214; 1,000 fathoms; Aug. 21.

DISTRIBUTION: This species, first described from the Malayan region, has subsequently been found in the Indian Ocean (Moser, 1925; Browne, 1926), on both sides of the Pacific, off the Great Barrier Reef of Australia (Totton, 1932), in the China Sea (Bigelow, 1913), in the eastern tropical Pacific (Bigelow, 1911), and also in the South Atlantic (Moser, 1925).

14. *Eudoxoides mitra* Huxley 1859.

MATERIAL: No. 311199; Net 1169; 700 fathoms; Aug. 12.

DISTRIBUTION: This species, first described from the Indian Ocean, is well known in the Pacific (Totton, 1932; Bigelow, 1911, 1913), and in the Atlantic (Moser, 1925; Leloup, 1933, 1934; Bigelow, 1918).

15. *Eudoxoides spiralis* Bigelow 1911.

MATERIAL: No. 311534; Net 1214; 1,000 fathoms; Aug. 21.

DISTRIBUTION: This species is cosmopolitan in warm seas; the records include the eastern tropical Pacific (Bigelow, 1911), Japanese waters (Bigelow, 1913), off the Great Barrier Reef of Australia (Totton, 1932), the Indian Ocean (Browne, 1926), many localities in the South Atlantic, south to Latitude 45° S. (Moser, 1925; Leloup, 1934); likewise the tropical and subtropical North Atlantic, northward to the vicinity of the Cape Verdes (Leloup, 1934) on the one side and in the region of Cape Hatteras, Bermuda and the Bahamas (Bigelow, 1918) on the other. It has also been taken in the Mediterranean.

16. *Lensia conoidea* Keferstein and Ehlers 1860.

MATERIAL: No. 3174; Net 988; 1,000 fathoms; June 2.
No. 311816; Net 1263; 800 fathoms; Sept. 4.

COLOR: Lemon yellow in life.

DISTRIBUTION: Since *L. fowleri* and *L. subtiloides* have often been treated as synonyms of *L. conoidea*, it is impossible to learn its range from published accounts. There are definite records of it from the North Pacific, the Malayan region, the Gulf Stream, and the coast of Norway. It is likely that it occurred among the specimens listed as "*truncata*" from the South Atlantic (Moser, 1925; Leloup, 1934; Hardy and Gunther, 1935), and in the Mediterranean (Moser, 1925; Leloup, 1933). It was taken in the latter sea in abundance by the *Thor* (Bigelow and Sears, in press).

17. *Lensia multicristata* Moser 1925.

MATERIAL: No. 311415; Net 1195; 800 fathoms; Aug. 17.
No. 311780; Net 1258; 900 fathoms; Sept. 3.

DISTRIBUTION: *Lensia multicristata* Moser is widespread in the eastern tropical Pacific (Bigelow, 1911), in the Indian Ocean (Browne, 1926), in the South Atlantic (Moser, 1925; Leloup, 1934), and as far north in the North Atlantic as the Bay of Biscay (Bigelow, 1911a), as well as in the Mediterranean (Bigelow and Sears, in press).

18. *Lensia profunda* sp. nov.[3]

MATERIAL: No. 311377; Net 1190; 900 fathoms; Aug. 16.
No. 311601; Net 1217; 600 fathoms; Aug. 24.

19. *Lensia* sp.

MATERIAL: No. 311534; Net 1214; 1,000 fathoms; Aug. 21.

20. *?Chuniphyes multidentata* Lens and Van Riemsdijk 1908.

MATERIAL: No. 311199; Net 1169; 700 fathoms; Aug. 12.
No. 311510; Net 1207; 900 fathoms; Aug. 20.
No. 311968; Net 1291; 600 fathoms; Sept. 12.
No. 312008; Net 1298; 800 fathoms; Sept. 14.

COLOR: Transparent and colorless in life.

DISTRIBUTION: The bathypelagic species, *C. multidentata*, was first described from Malayan waters. Later, it has been recorded from the eastern tropical Pacific (Bigelow, 1911, 1931), the offing of California (Bigelow and Leslie, 1930), the Eastern Sea between Japan and China (Bigelow, 1913), near the Philippines (Bigelow, 1919), and on both sides of the Atlantic—south to South Georgia, and north to the Bay of Biscay (Bigelow, 1911a; Moser, 1925; Leloup, 1933, 1934; Hardy and Gunther, 1935). There is only one record of it within the Mediterranean (Leloup, 1933).

Suborder Physophorae.
Family Forskalidae.
21. *Forskalia* sp.

MATERIAL: No. 3110; Net 980; Surface; May 17.
No. 311300; Net 1185; 900 fathoms; Aug. 15.

[3] See MS. *Discovery* Report.

No. 311301; Net 1185; 900 fathoms; Aug. 15.
No. 311511; Net 1207; 900 fathoms; Aug. 20.

COLOR: No. 311 0 was described as "lemon-colored," No. 311300 as "orange-red," and No. 311511 as "black-lined."

Family Agalmidae.

22. *Agalma elegans* Sars 1846.

MATERIAL: No. 31492; Net 1054; 25 fathoms; July 7.
No. 312117; Net 1314; 600 fathoms; Sept. 7.

COLOR: Transparent and colorless.

DISTRIBUTION: This species is well known from the Mediterranean, from the coasts of Europe as far north as Norway and along the eastern coast of North America from Cape Cod to the West Indies, and in the easttern tropical Pacific (Bigelow, 1911), as well as in Malayan waters (Bedot, 1896) and the Indian Ocean (Browne, 1926).

23. *Agalma okeni* Eschscholtz 1825.

MATERIAL: No. 31210; Net 1018; 900 fathoms; June 15.
No. 31314; Net 1038; 300 fathoms; June 25.
No. 31976; Net 1128; 400 fathoms; Aug. 4.
No. 311616; Net 1226; 300 fathoms; Aug. 27.
No. 312068; Net 1308; 100 fathoms; Sept. 16.
No. 312071; Net 1308; 100 fathoms; Sept 16.
No. 312085; Net 1310; 300 fathoms; Sept. 16.
No. 312088; Net 1311; 300 fathoms; Sept. 16.
No. 312170; Net 1322; 300 fathoms; Sept. 18.

COLOR: Nectophores transparent, siphosomes coral and white in life.

DISTRIBUTION: This species occurs in the warmer regions of all the great oceans and has occasionally been taken in the Mediterranean and Red Sea.

24. *Stephanomia amphitridis* Péron and Lesueur 1807.

MATERIAL: No. 3152; Net 984; 600 fathoms; June 2.
No. 3153; Net 984; 600 fathoms; June 2.
No. 311013; Net 1133; 700 fathoms; Aug. 5.
No. 311040; Net 1133; 700 fathoms; Aug. 5.
No. 311091; Net 1143; 500 fathoms; Aug. 7.
No. 311092; Net 1143; 500 fathoms; Aug. 7.
No. 311112; Net 1149; 500 fathoms; Aug. 8.
No. 311534; Net 1214; 1,000 fathoms; Aug. 21.
No. 311859; Net 1274; 900 fathoms; Sept. 7.
No. 312196; Net 1326; 600 fathoms; Sept. 19.

COLOR: Varying from crimson to pink and orange.

DISTRIBUTION: This species, originally described from the Atlantic, has also been recorded from the Pacific, the Malayan region and probably from Ceylon.

Subfamily Nectaliinae.

25. *Nectalia loligo* Haeckel 1888.

MATERIAL: No. 311040; Net 1133; 700 fathoms; Aug. 5.
No. 311113; Net 1149; 500 fathoms; Aug. 8.

No. 311494; Net 1205; 700 fathoms; Aug. 20.
No. 311570; Net 1218; 700 fathoms; Aug. 24.
No. 311571; Net 1218; 700 fathoms; Aug. 24.
No. 311673; Net 1236; 600 fathoms; Aug. 29.
No. 311688; Net 1243; 700 fathoms; Aug. 31.
No. 311968; Net 1291; 600 fathoms; Sept. 12.
No. 311984; Net 1291; 600 fathoms; Sept. 12.
No. 311964; Net 1292; 700 fathoms; Sept. 12.
No. 311966; Net 1292; 700 fathoms; Sept. 12.

COLOR: Crimson and transparent.

DISTRIBUTION: Few specimens of this species are on record. The type came from the Canary Islands and subsequent records are two specimens taken by the Plankton Expedition in 3° 6′ N. Lat., 33° 2′ W. Long., and the northern border of the Gulf Stream south of Iceland, one from Orotava, one from the eastern tropical Pacific, and from the South Atlantic (Moser, 1925).

Subfamily Anthophysidae.

26. *Anthophysa formosa* Fewkes 1882.

MATERIAL: No. 311187; Net 1162; 800 fathoms; Aug. 11.
No. 311281; Net 1178; 900 fathoms; Aug. 14.
No. 311367; Net 1188; 500 fathoms; Aug. 16.
No. 312205; Net 1325; 500 fathoms; Sept. 19.

COLOR: Lemon yellow and white.

DISTRIBUTION: This is the member of this genus found in the Atlantic, and may eventually prove to be identical with *A. rosea* Brandt. It is known from the Gulf Stream in the western side of the North Atlantic, from the Sargasso Sea, from the South Atlantic, where it was taken by the *Challenger,* from the vicinity of the Canaries and from the Mediterranean.

27. *Athorybia rosacea* (Forskål 1775) Eschscholtz 1829.

MATERIAL: No. 311008; Net 1136; 1,000 fathoms; Aug. 5.

28. *Athorybia* sp.

MATERIAL: No. 312071; Net 1308; 100 fathoms; Sept. 16.

Suborder Rhizophysaliae.

Family Rhizophysidae.

29. *Rhizophysa* sp.

MATERIAL: No. 31719; Net 1084; 25 fathoms; July 15.

COLOR: Pink.

Family Physalidae.

30. *Physalia physalis* Linné 1758.

MATERIAL: Often taken at the surface.

DISTRIBUTION: This is the common Portuguese-man-of-war of the warmer parts of the Atlantic Ocean.

Suborder Chondrophorae.

Family Porpitidae.

31. *Porpita umbella* O. F. Müller 1776.

MATERIAL: Occasionally taken at the surface.

DISTRIBUTION: This species is the one found in the warm waters of the Atlantic Ocean. It may prove to be the same as *P. porpita* Linné from the Indian Ocean, but it is probably distinct from *P. pacifica* Lesson.

Family Velellidae.

32. *Velella velella* Linné 1775.

MATERIAL: Occasionally taken at the surface.

DISTRIBUTION: This species is found in the warmer waters of the Atlantic and Pacific. On further study, it may prove identical with the *Velella* of the Indian Ocean.

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21.

The Female Bitterling as a Biologic Test Animal for Male Hormone[1].

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(Plate I; Text-figure 1).

INTRODUCTION.

A number of different methods of testing for male hormone have been proposed (1) with the capon, the guinea pig, the rat, and the castrated male bitterling (2) as test animals. None of these tests has been very satisfactory. Recently Witschi (3) has described a change in the color of the bill of the sparrow after injections of male hormone and has suggested this as an indicator. The capon test is the one most commonly employed. Injections of potent material cause a growth of comb and wattles in four or five days. The measurement is not simple nor is it standardized (4). Injections do not produce constant results and for each test six animals should be used (5). Thus a considerable amount of expense, space, care, and attention are required for a single test. Castration must be complete and the bird can not be used again after a positive reaction until the comb has regressed.

It therefore seems evident that a more convenient method of detecting and measuring the male hormone would be of considerable value. The ovipositor lengthening of the female bitterling appears to furnish a simple biological reaction for this hormone, as the present experiments will indicate. This lengthening of the ovipositor of the female bitterling (Pl. I, Fig. 1) was first elicited by Ehrhardt and Kuhn (6) and, shortly after, independently by Fleischmann and Kann (7), but they ascribed this reaction to follicular hormones. It was later suggested as of possible use in diagnosing pregnancy by Kanter, Bauer, and Klawans (8). Although these authors did not definitely state that it was a test for pregnancy, their paper intimated this use so clearly that a number of laboratories accepted this viewpoint. It was soon shown that it could not be so considered (9, 10, 11), since urines from non-pregnant females, women in the post-climacteric period, and even from men gave positive reactions. Occasionally a pregnant women's urine would be negative. That this test is produced by male hormone will be shown below (12).

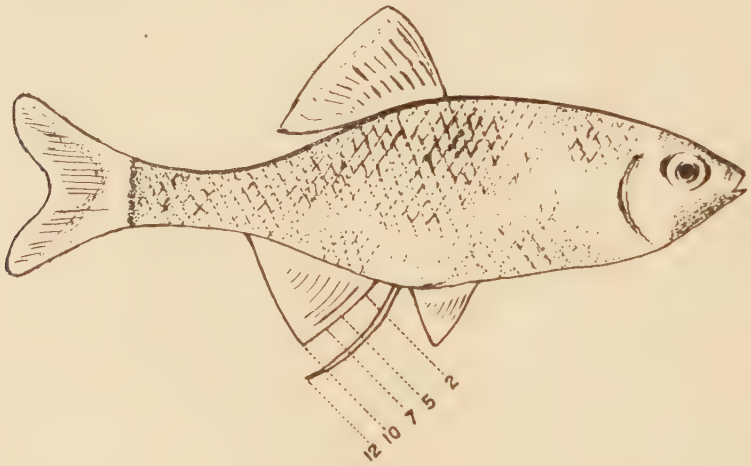
[1] Aided by a grant from the Lucius N. Littauer Foundation.

Experimental.

The female bitterling[2] develops an ovipositor which depends from the ventral margin of the body slightly anterior to the origin of the anal fin. In the quiescent state the ovipositor is seldom visible, but in those individuals in which it is visible out of season it is very small and rarely reaches 5 mm. in length. During breeding activity the organ is prolonged until it may reach 5 cm. in length (Pl. I, Fig. 2) and at spawning it is inserted into the inhalent siphon of a mussel, usually *Unio or Anodonta,* and the ova extruded into the gill-folds. Fertilization is effected by sperm which is liberated near and drawn into the inhalent siphon of the mussel and passed over the embedded ova. Hatching occurs within the gill-folds and the fry liberate themselves, in a post-larval state, two or three weeks after oviposition.

Description of Test.

To a small aquarium is added 4 liters of water, 2 liters from a stock tank and 2 liters from the tap. Two female bitterlings are placed in the tank and kept there 24 hours before introducing the material to be tested. At the end of the 24-hour period readings are made of the size of the ovipositor. The scale used is as follows: If the ovipositor is not visible the reading is 0; if the length of the ovipositor equals the length of the first ray of the anal fin the reading is 10; an ovipositor which reaches halfway down the first ray is 5; etc. (That is, the length of the ovipositor is compared to that of the first ray of the anal fin in equal units running from 0 to 10. (Text-fig. 1).) No fish with ovipositors exceeding 3 on the scale, before the addition of any test material, are used.



Text-figure 1.

Diagrammatic representation of scale used in reading length of ovipositor of female bitterling.

Ovipositor readings are taken at 24, 48, and 72 hour intervals and maximum growths under these conditions are usually observed at the second reading.

We tentatively define a bitterling unit as the amount of material which, when added to a tank containing 4 liters of water and 2 female bitterlings,

[2] We are greatly indebted to Mr. C. M. Breder, Jr., of the New York Aquarium, for advice and suggestions in respect to the fish.

produces an increase in the length of the ovipositor of one or both fish of 7 or more on the scale within 48 hours.[3] For assay it is suggested that a series of dilutions may be set up and the lowest dilution giving a positive reading may be considered to contain at least one unit, providing a positive reaction is also given in the next higher dilution.

In this work the European bitterling (*Rhodeus amarus*) was used. Kanter et al (8) used the Japanese bitterling (*Acheilognathus intermedium*) and suggest differences between the two species. However, comparative initial experiments with pregnancy urines conducted under identical conditions at the same time with both species by Coates (unpublished) did not seem to indicate any substantial difference between them.

An important observation is that female bitterlings which have been used throughout the year seem quite refractory during the late spring and summer months. From about May 15 until August 15 they react very weakly, as a rule, to preparations which at other times are definitely effective. This has been noted by other investigators. It may be a temperature effect. Gottlieb (11) working in Quebec, got good results throughout the breeding season, i. e., May-July. Whether temperature regulation or some other means may be devised to overcome this difficulty remains to be seen. In the meantime, this seasonal variation must be taken into account.

The preliminary experiments (9) already referred to, showed that urines from non-pregnant females, pregnant females, women in post-climacteric period, and men gave positive reactions in many instances. We have since extended these observations and these indicate that most normal urines from young adult males cause reactions if a large enough quantity can be used without harm to the fish. This suggested that the responsible factor might not be the follicular hormones. Moreover, the fact that urines from women 5-6 years after menopause still gave ovipositor reactions in some cases gave further support to this hypothesis. We also conducted a large number of experiments with commercial medicinal female sex hormone preparations and obtained very irregular results. Up to this point the results did not definitely indicate that the responsible factor is hormonal in nature. It might possibly be some common urinary constituent, such as creatinine, uric acid, or indican.

It was therefore seen to be necessary to determine what type of compound present in urine caused the reaction. Urine was first dialysed with the following results:

Experiment 1: Urine was obtained from a healthy male subject whose urines, previously tested, had invariably given positive results. 20 cc. was dialysed in a cellophane membrane against 40 cc. of distilled water in the refrigerator, for 24 hours. An undialysed portion was kept at the same temperature for the same period. 16 cc. of the dialysed urine, 32 cc. of the dialysate, and 16 cc. of the untreated urine were each added to 4 liters of water containing 2 fish. Positive reactions were noted in 48 hours in the tanks to which had been added the dialysed urine, as well as the control, but not in that containing the dialysate.

Experiment 2: A similar experiment was done with pregnancy urine. Similar results were obtained but it was noted that the dialysed urine was distinctly ammoniacal. This led to the suspicion that an excess of ammonium compounds might be the causative factor. An experiment was therefore conducted in which ammonium hydroxide was added to the water containing 2 fish. Results of this test proved to be negative. It thus seemed evident that the ovipositor stimulating substance is not an ammonium salt and is not dialysable. This also rules out many common urinary constituents, such as inorganic salts, creatinine, urea, uric acid, etc.

[3] This differs slightly from our originally suggested definition (12), i.e., that the ovipositor must reach the end of the fin. The present definition allows for differences in the initial length of the ovipositor.

The next step was to determine whether it belonged to any of the classes of sex hormones. Even though we had shown its incapability of being used as a pregnancy indicator, it was necessary to rule out definitely the anterior-pituitary-like hormone. This was easily done by boiling some "positive" urine and ascertaining that the resulting urine was still positive. The anterior-pituitary-like hormone ("AP-L") is heat labile.

The following experiment showed that the responsible factor is chloroform-soluble.

Experiment 3: 500 cc. of mixed urine from 20 pregnant women was treated with 20 cc. of conc. HCl and was then extracted three times with 100 cc. portions of chloroform in a separatory funnel. An emulsion formed which was cleared by the addition of 500 cc. of ether. The chloroform-ether layer was drawn off, evaporated on a steam bath, and the brown gummy residue dissolved as far as possible in 65 cc. of water containing 0.5 cc. of 10% NaOH. This was added to 7 liters of water containing 2 female bitterlings. In 16 hours both fish showed marked positive reactions. 35 cc. of the extracted urine from which the chloroform had been removed gave no reaction under similar conditions in 72 hours. This experiment led us to the conclusion that the effective factor is soluble in chloroform and to the impression that it is similar to the ovarian follicular hormones.

It therefore seemed logical to expect that crystalline "theelin" or "theelol," or both, would produce this reaction. We were fortunately able to obtain a small amount of each from Dr. Edward A. Doisy and they were tested and showed very slight effects, if any, on the ovipositor.

As is well known, the chloroform and ether soluble hormones of urine comprise the male as well as the female sex hormones, i. e., androsterone and estrins. We consequently subjected urine to a rough separation of these substances, using the method of Funk, Harrow, and Lejwa (13) with the modifications of Butenandt and Tscherning (14) and Kochakian and Murlin (15). A typical experiment is the following:

Experiment 4: 18 liters of mixed male urine was made acid to Congo Red and 360 cc. conc. HCl added. The mixture was concentrated on a steam bath to 1,800 cc. 500 cc. chloroform was added and the mixture refluxed for 12 hours on a steam bath. The aqueous fraction was discarded and the chloroform extract evaporated to dryness. The gummy residue was then dissolved in ether and shaken with 2N KOH until no further color appeared in the aqueous phase. The washings were extracted with ether and the ether solutions combined, and evaporated to dryness. The residue was refluxed with 50 cc. 3N KOH in methyl alcohol for 2 hours. After cooling, 2 liters of water was added and extracted repeatedly with ether. The aqueous fraction was saved. The extract was washed with water, dilute acid, and finally again with water. This constitutes the male hormone fraction.

The aqueous fraction was treated with HCl until acid to Congo Red and an additional 20 cc. HCl were added. The mixture was heated on a steam bath for 1 hour and extracted repeatedly with ether. The ether extract was washed with water, dilute alkali, and finally with water. This yields the female hormone fraction.

Each fraction was now tested for its ovipositor-lengthening effect on the female bitterling, as follows:

Experiment 5: Ether solutions equivalent to 64 cc. of urine were placed in a mortar and the ether permitted to evaporate. The oily residue in each case was emulsified with acacia and water and added to a tank of 2 bitterlings in the usual manner. After 18 hours positive reactions were seen to have been produced by the *male fraction* and none by the *female fraction.*

That the *male fraction* actually contained male hormone was substantiated by injection of a cotton-seed oil solution of it into a capon. A posi-

tive result was seen, whereas a similar test with the female fraction was negative. Both fractions, however, produced estrogenic effects when injected into immature female mice. This harmonizes with the previous experiences of many investigators (16) who found estrogenic effects with male hormone preparations. Recent work indicates, however, that highly purified or synthetic androsterone is non-estrogenic by the vaginal cornification test (17). Hence, the estrogenic effect of our male fraction may have been due to admixture of impurities.

The question now arose as to why pregnancy urines should be more potent and more constant in their activity than urines from males and non-pregnant females. We therefore repeated the above experiment, using mixed pregnancy urines. Here again the bitterling test was positive with the male fraction and negative with the female.

The facts thus indicate that the ovipositor-lengthening factor of urine is present in the male fraction, but proof that one of the male hormones is responsible was still lacking. This gap in the proof has recently been filled by the use of *synthetic* preparations[4]. Despite the fact that our experiments were performed during the refractory season we were successful in showing the efficacy of these products. At the same time we can also report what seems to be a more suitable solvent which is harmless to the fish and which offers a much better menstruum for these sterols than the acacia which we had formerly employed as an emulsifying agent. This is propylene glycol, suggested to us by Dr. Warren M. Cox, Jr., of the Research Laboratory of Mead Johnson & Co.

Experiment 6: 4 mg. of synthetic androsterone (Schering) and 3 mg. of synthetic testosterone (Schering) were each dissolved in 5 cc. of propylene glycol (Eastman) under the influence of slight heating. Aquaria with two bitterlings in 4 liters of water had been set up the previous day. All had extremely small ovipositors (0 to 2). One liter of water was removed from the aquarium and the propylene glycol solution of the sterol added quickly and shaken vigorously. It was then added to the aquarium from which the water had been taken. A third aquarium with the same amount of propylene glycol was observed as a control.

A positive result was noted in the bitterlings exposed to both the crystalline androsterone and testosterone. Propylene glycol alone was negative. The fish exposed to androsterone were greatly weakened by this substance but the testosterone had very little systemic effect.

In the fall when the animals were found to be in a normally reactive state both of these synthetic products were re-tested. It was found that 0.8 to 1.2 mg. of androsterone (18) produced positive reactions in 48-72 hours, whereas small amounts were never effective and larger doses yielded variable results. The larger doses seemed to have the same depressing effect as was mentioned above. Crystalline testosterone also gave uniformly positive reactions at a certain dosage (0.6-0.8 mg.) (unpublished data) whereas larger and smaller amounts were usually ineffective. Again it may be stated that a positive reaction is one in which the lengthening of the ovipositor totals 7 points or more on the scale. Many of the negative tests at other dosages showed slight effects, i. e., a lengthening of less than 7. It must also be pointed out that the results with these crystalline products appeared more slowly than when urine had been used, presumably because they are in a different physical state. From these experiments *it is evident that the ovipositor-lengthening phenomenon is due to male hormones.* There can be no suspicion of admixture of urinary impurities in these synthetic products.

Confirmatory evidence has also been obtained by testing male hormone concentrates kindly furnished by Dr. Benjamin Harrow of the College of

[4] We wish to thank Schering and Company and Dr. Erwin Schwenk for the synthetic androsterone and testosterone supplied.

the City of New York. This material, an oily solution of the male hormone fraction of male urine, has an activity of 10 capon units per cc., each cubic centimeter representing 1,000 to 1,150 cc. of urine. As little as 0.1 cc. added to an aquarium of female bitterlings gives a positive reaction in 48 hours. The minimum effective dose has not yet been reached.

Technique for Assay of Urine.

In attempting to determine the amount of male hormone in normal male urine, the following procedure was employed, using normal male medical students as subjects: For each assay, 4 aquaria were set up, containing 4 liters of water and 2 female bitterlings in the usual manner. To each tank was then added 10 cc., 25 cc., 50 cc., and 100 cc. portions of urine respectively. 24 hour samples of urine were used. The smallest amount causing a positive reaction was considered to contain one unit. The number of units in a 24-hour sample was then calculated. It was noted that in many instances the urine was toxic and even fatal in amounts from 50 to 100 cc. Various methods have been tried in the attempt to detoxicate the urines and finally it was found that dialysing is all that is necessary (19). By this means dialysed urines representing as much as 200 cc. of the original may be used without any ill effects whatever. Ordinarily the procedure is the following: Measured amounts of urine, usually 200 cc., are dialysed in membranes of cellophane ("plain transparent" not "moisture-proof") against running tap water for 18-24 hours. The volumes are then measured and amounts equivalent to 10, 25, 50, and 100 cc., respectively, of the original urine are added to 4 aquaria, each of which contains 2 female bitterlings. The ovipositors of the fish must have been read on the preceding day and also just before addition of the dialysed urine. Subsequent readings are made in 24, 48, and 72 hours, and the number of units determined in the manner suggested above. The average excretion of male hormone is about 35 bitterling units per day with a range of approximately 15 to 75 b.u. (see Table I). Further work to check and enlarge this series is in progress.

TABLE I.

Daily excretion of male hormone by normal adult males.

Number of 24-hour urine specimens	Number of cc. of urine which, when added to 4 liters of water, produce a positive reaction	Bitterling units excreted daily
5	10—15	75—120
15	25—35	24—45
10	75—100	9—22

Preliminary tests have indicated the unreliability of using casual specimens for even rough quantitative work. We have noted that successive samples obtained during the day from the same individuals have been exceedingly variable in the amounts of hormone excreted and some samples are even entirely devoid of the hormone. Apparently 24-hour samples of urine are needed in order to determine the output of this hormone with any degree of accuracy.

Discussion.

Evidently neither theelin nor theelol is responsible for the reaction, nor are various cholane derivatives which have thus far been tested, such as cholesterol, ergosterol, and sodium taurocholate (20). Several of these

give slight reactions, which suggest the possibility that other cholane derivatives may be found which will react as well as the male hormone. Up to the present the test seems to be specific for male hormone.

In support of this we may cite several references to the literature. Glaser and Haempel (21) compared the effect of a follicular hormone preparation with several other preparations, including a male hormone product, "testosan forte." The technique of the experiments included placing males and females in the same aquarium. The results showed slight growth of ovipositors in those females subjected to the male as well as the female hormone, although the latter gave stronger results. The data given seem inconclusive, however. Glaser and Haempel conclude that certain secondary sex characteristics are influenced by both male and female hormones. Among these are the lengthening of the ovipositor of the female bitterling and the growth of the comb and wattles of the capon. This work was subjected to criticism by Fleischmann and Kann (22). They confirmed Glaser and Haempel's contention that "testosan forte" gave positive results with the female bitterling but they are of the opinion that this is due to an admixure of follicular hormone. In support of this they state that this male hormone preparation also gives the Allen-Doisy test on the mouse. A more concentrated male hormone preparation, "Proviron" (Schering) had the same action, i.e., positive reactions with both the fish and mouse tests, whereas crystalline "Proviron" reacted negatively with both. We suggest that this negative result may have been a result of incomplete solution or improper emulsification of the substance. The authors do not state the menstruum or method employed for this purpose.

An interesting contribution has also been made by Ehrhardt and Kuhn (23). In a long series of experiments they come to the conclusion that the ovipositor-lengthening factor and the estrus hormones are not identical, although both have a number of properties in common, i.e., solubility in organic solvents, heat stability, absorption on charcoal, etc. Some points of difference are the following: in the urine of pregnant mares are large quantities of estrus hormone, but small quantities of the ovipositor-lengthening factor. Non-pregnant women, on the other hand, excrete a urine with just the opposite characteristics. Eight hundred units of technical (i.e., impure) estrus hormone give a better ovipositor-lengthening result than several thousand units of crystalline estrus hormone. The blood serum of the pregnant woman gives a strong Allen-Doisy test but a weak bitterling test. Their preparations thus did not quantitatively show parallel results when tested with the fish and with the mouse. Although they believe the two phenomena to be due to two distinct substances they are of the opinion that one is a transformation product of the other. Ehrhardt and Kuhn's work harmonize with the contention that the male hormone is the responsible factor for this phenomenon.

We propose to assay urines and other biological fluids under various physiological conditions and from pathological cases for their male hormone content by this method. The method should also be of value in assaying male hormone products to be used therapeutically.

Summary.

The lengthening of the ovipositor of the female bitterling produced by the administration of human urine has been found to be due to the male hormone present therein. Confirmation of this is shown by positive effects caused by crystalline androsterone and testosterone. A bitterling unit is provisionally defined and a method of measuring the ovipositor is described.

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EXPLANATION OF THE PLATE.

Plate I.

Fig. 1. Female bitterling, showing elongation of ovipositor: considered a minimum positive reaction.

Fig. 2. Female bitterling, showing elongation of the ovipositor in the process of natural ovipositation.

FIG. 1

FIG. 2

THE FEMALE BITTERLING AS A BIOLOGIC TEST ANIMAL FOR MALE HORMONE.

22.

Some Tropical Fishes as Hosts for the Metacercaria of *Clinostomum complanatum* (Rud. 1814) (= *C. marginatum* Rud. 1819).

Ross F. Nigrelli.
New York Aquarium.

(Plates I & II).

The following fishes received by the New York Aquarium were found infected with the yellow grub stage of *Clinostomum*: *Chriopeops goodei* (Jordan) from Florida; *Mollienisia velifera* (Regan) from Yucatan; *Piabucana* sp. from the Amazon drainage; *Corynopoma riisei* Gill from British Guiana; *Nannostomus trifasciatus* Steindachner from the Tucantins, a tributary of the Amazon; *Lebistes reticulatus* (Peters) from out-door pools in Florida; *Hypopomus artedi* Kaup from the Amazon drainage in eastern Brazil; and *Sternopygus macrurus* (Bloch and Schneider) from the Amazon drainage.

In some instances the fishes showed no infection when they arrived, and only after they had been in the Aquarium for some time did the cysts appear. Whether the fishes were originally infected and growth of the parasite had taken place in the tanks, or whether the fishes acquired the infection in the tanks is difficult to determine. Examination of snails and water of the aquaria showed no fork-tailed cercariae.

In cases of severe infection the death of the host resulted. The trematodes were usually encysted in the fish, and in *Corynopoma riisei* the grub was embedded deep in the peculiar transparent muscle of the body (Pl. I, Figs. 1 & 2). In the gymnotid, *Sternopygus macrurus,* 16 cysts were found distributed along both sides of the base of the elongated ventral fin.

Observations on the "guppy"' showed that, since this species is not influenced by seasonal changes, the cysts are not deserted as reported by Ward (1918) for northern forms. The parasites were present on the fish (Pl. II, Fig. 3) for more than a year. In one of the specimens found dead, moribund metacercariae were still in the cysts. Many of the temperate zone fishes in the Aquarium were also found infected with the metacercariae of *Clinostomum.* In the majority of instances the worms were encysted on the membrane between the rays of the fins. These cysts appeared in the spring and in winter were absent.

Examination of the parasites showed no morphological differences between the metacercariae found on tropical forms and those of temperate zone species. In all instances the worms have been identified as *C. complanatum* (Rud. 1814) (= *C. marginatum* Rud. 1819) (Pl. II, Figs. 4 & 5).

The metacercaria of *C. complanatum* has been found on a large number of fish hosts throughout the world. As *C. marginatum* it has been reported by several investigators from the following neotropical fishes (see Baer 1933): *Adnia* (*Adinia?*) *dugesi* (Bean), *Callichthys asper* Quoy and

Gaim., *Poecilia vivipara* Bloch and Schneider, *Chaetosomus brachyurus* (Kner), *Cynodon scomberoides* (Cuv.), *Satanoperca papatera* (Heck.), *Chaetobranchus flavescens* Heck., *C. gulosus* (Cuv. and Val.), *Crenicichla johanna* Heck., *Crenicichla saxatilis* (Linn.).

These are definitely Central and South American species of fishes, but because of the terminology employed, it is difficult to determine which form is referred to, since in the present day usage some of the above genera have been split up and others have been discarded. In North America the metacercaria has been reported from a large number of fishes, several species of amphibians (*Rana clamitans, R. pipiens,* etc.), and, according to Hopkins (1933), the parasite was found in a snake (*Thamnophis radix* (Baird)) by van Cleave.

The following are some of the North American fishes found infected: *Perca flavescens* (Mitchill)[1] (yellow perch); *Aphredoderus sayanus* (Gilliams) (pirate perch); *Micropterus dolomieu* Lacépède[1] (small mouthed black bass); *M. salmoides* (Lac.)[1] (large mouthed black bass); *Ambloplites rupestris* (Raf.)[1] (rock bass); *Lepomis pallidus* (Mitchill)[1] (bluegilled sunfish); *L. auritus* (Linn.)[1] (red-breasted sunfish); *L. cyanellus* Raf.[1] (green sunfish); *Eupomotis gibbosus* (Linn.)[1] (common sunfish); *Catostomus commersoni* (Lac.)[1] (common sucker); *Pimephales promelas* Raf. (fathead minnow); *Semotilus astromaculatus* (Mitch.) (horned dace); *Ameiurus nebulosus* (Le Sueur)[1] (common bullhead); and *Salvelinus fontinalis* (Mitch.) (brook trout). In Europe Ciurea (1911) reported the yellow grub of *C. complanatum* on *Perca fluviatilis* Linn., and Maccagno (1933) found it on the loach, *Cobitis taenia* Linn. In Japan Yamaguti (1933) found it under the skin and flesh of *Carassius carassius* Linn., *Pseudogobio esocinus,* and *Acheilognathis intermedia.*

The following are other species of *Clinostomum* described from fishes:

1. *C. heterostomum* (Rud. 1809) from European fishes, and from an unidentified species taken in the Vaal river, Africa (Monnig 1926).
2. *C. dimorphum* (Diesing 1850) from many of the tropical fishes mentioned above.
3. *C. dictyotum* (Monticelli 1893) (= *Clinostomatopsis reticulata* (Looss 1885)) from an unidentified silurid of Costa Rica.
4. *C. africanus* Stoss. from the intestine of an unidentified fish of the French Congo (see Galli-Valerio 1906).
5. *C. piscidium* Southwell and Prashad 1918 from *Trichogaster fasciatus* and *Nandus nandus* of Ceylon, and from *Trichogaster pectoralis* and *T. trichopterus* of Siam (Pearse 1933).
6. *C. intermedialis* Lamont 1920 from the silurid, *Rhamdia quelen* Quoy and Gaimard.
7. *C. chrysichthys* Dubois 1930 from *Chrysichthys kingselyi* Gunth., a silurid of Angola.
8. *C. clarias* Dubois 1930 from *Clarias angolensis* Steind., a silurid of Angola.
9. *C. dalgi* Tubangiu 1933 from the eye-socket and pericardium of *Ophiocephalus striatus* Bloch., a Philippine fish.

Four orders of fishes are represented by the hosts of *Clinostomum* reported in the literature. Arranged according to the number of species found infected, these are: Ostariophysii, Acanthopterygii, Haplomi, and Isospondyli. With the exception of the Isospondyli, all are basically pond fishes. In the great majority of ponds the same relationship among these orders probably exists; that is, the Ostariophysii includes the largest number of species to be found in such a habitat, the Acanthopterygii the next largest, etc.; and likewise as to number of individuals. The Isospondyli is repre-

[1] Species present in the Aquarium on which *Clinostomum* was found.

sented by one species, *Salvelinus fontinalis* (brook trout). It is altogether possible that this form became infected during an excursion into still water, which it sometimes makes.

It is not surprising to find that pond fishes show the highest incidence of infection. The nature of the cercaria of *Clinostomum* is such that it would be mechanically difficult to infect fishes in a fast stream. These cercariae are fork-tailed organisms which float at the surface. The pond fishes, moreover, are more sluggish than the stream forms, thus making it more feasible for the cercariae to penetrate the skin.

The genus *Clinostomum* was erected by Leidy (1856) for a metacercaria found encysted in the skin of the common sunfish, *Pomotis vulgaris* C. and V., *Eupomotis gibbosus* (Linn.), and in the intestine of *Esox* sp. This species he called *C. gracile* and designated it as the type of the genus. *C. gracile* Leidy 1856, however, is considered a synonym of *C. complanatum* (Rud. 1814), an adult form taken from a heron (*Ardea cinerea*) by Rudolphi and described by him as *Distomum complanatum*. Braun (1901) made *C. complanatum* the type of the genus because Leidy's description was inadequate and because the original type was lost. There is no doubt, however, that the yellow grub, called *C. marginatum* by most North American investigators and even now found on the type host and in the type locality, is Leidy's *C. gracile*. Braun (1899) considered *C. marginatum* (Rud. 1819) a synonym of *C. complanatum* (Rud. 1814) because the two were morphologically the same and were found as metacercaria on fishes and as adults in the same genus of birds (*Ardea*). Baer (1933) expressed the same opinion and added that the geographical distribution of the two was alone not sufficient to warrent regarding them as distinct species.

According to Baer (1933) *C. heterostomum* (Rud. 1809) = *Euclinostomum heterostomum* (Rud. 1809), and *C. dimorphum* (Diesing 1850) = *Ithyclinostomum dimorphum* (Diesing 1850). In *Euclinostomum* Travassos 1928 the intestinal branches have long lateral diverticula, sometimes ramifying. In *Ithyclinostomum* Witenberg 1926 the lateral diverticula are short and never ramifying. This genus is further separated from *Clinostomum* on the distribution of the vitelline glands. In the latter these glands extend into the anterior part of the body.

The complete life history of *C. complanatum* was only recently worked out. It was known for some time that the adult worms could be found in the oral cavity of herons (*Ardea*) and related birds (Osborn 1911, 1912). In this country Hunter and Hunter (1934, 1935) showed that when the eggs of the parasites drop from the mouth cavity of the definitive host into water, they hatch into miracidia which penetrate the liver of snails. Two gastropods, *Helisoma campanulatum* (Say) and *H. antrosum* (Conrad), were experimentally infected. In the snail each miracidium develops into a sporocyst which produces rediae. The rediae in turn produce daughter rediae which become fork-tailed cercariae. These emerge from the snail and encyst as metacercariae, commonly called yellow grubs, on fishes. When an infected fish is eaten by a bird, the larvae are released and grow to maturity in the oral cavity.

Of the several species of *Clinostomum* described from fishes, the only other forms in which the adults are definitely known are *C. heterostomum* and *C. dimorphum* from ardeiform birds, and *C. intermedialis* from the cormorant (*Phalacrorax vigua*). Without doubt, however, a life-cycle similar to that of *C. complanatum* may be expected in the entire genus.

The following are species of adult *Clinostomum*, described from various definitive hosts, of which neither the first nor the second developmental stages are known: *C. dubium* Leidy 1856, *C. detruncatum* Braun 1899, *C. foliiforme* Braun 1899, *C. heluans* Braun 1899, *C. lambitans* Braun 1899, *C. sorbens* Braun 1899, *C. hornum* Nicoll 1914, *C. australiense* Johnston

1916, *C. pusillum* Lutz 1928, *C. phalacrocoracis* Dubois 1931, and *C. lophophallum* Baer 1933.

Two species of *Clinostomum* have been described from amphibians. Cort (1913) believed that the *C. marginatum* reported from batrachians (Wright 1897; Osborn 1911, 1912) was a distinct species, which he called *C. attenuatum*. His separation is based principally on certain morphological characteristics. He designated the American bittern as the probable definitive host of the metacercariae in amphibians, but Alvey and Stunkard (1932) reported *C. attenuatum* from the great blue heron. Hunter and Hunter (1934) demonstrated in feeding experiments that the great blue heron could be infected with metacercariae taken from frogs, but that the rate of infection was not as high as with *C. marginatum* from fish. They further showed (1935 a, b) that when both sunfish and tadpoles were exposed to the cercariae of *C. marginatum*, the sunfish became infected while the tadpoles remained free of metacercariae. They concluded that *C. attenuatum* is a distinct species. The other *Clinostomum* reported in amphibia is *C. pseudoheterostomum* Tubangui 1933, which occurs in the thigh muscle of a Philippine frog (*Rana magna*). The adult of this species, however, is unknown.

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EXPLANATION OF THE PLATES.

Plate I.

Fig. 1. *Corynopoma riisei* Gill showing the yellow grub encysted in the transparent muscle tissue. The worm appears as a dark spot behind the visceral mass.

Fig. 2. Fish shown in Fig. 1, immediately after death. The cyst was broken open and the living worm removed.

Plate II.

Fig. 3. Female "guppy", *Lebistes reticulatus* (Peters), about 1½ inches long, showing five or more *Clinostomum* cysts. Smaller males were also found infected.

Fig. 4. *Clinostomum complanatum* (Rud. 1814) from the "guppy" shown in Fig. 3. Enlarged about 22X.

Fig. 5. *Clinostomum complanatum* from the gymnotid, *Sternopygus macrurus*. Enlarged about 22X.

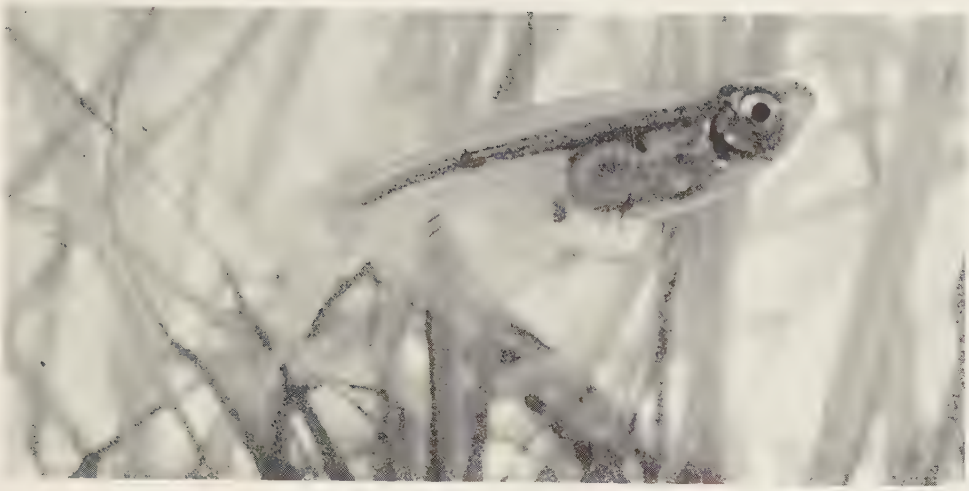

FIG. 1

FIG. 2

SOME TROPICAL FISHES AS HOSTS FOR METACERCARIA OF CLINOSTOMUM COMPLANATUM (RUD. 1814) (=C. MARGINATUM RUD. 1819).

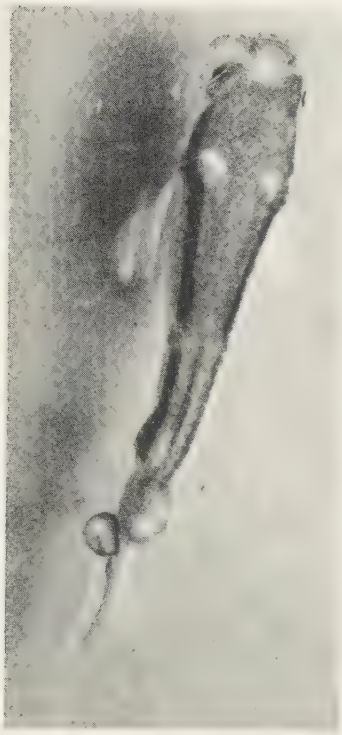

FIG. 3

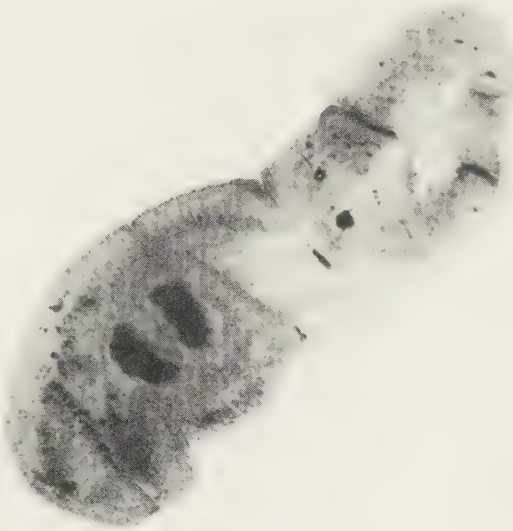

FIG. 4

FIG. 5

SOME TROPICAL FISHES AS HOSTS FOR METACERCARIA OF CLINOSTOMUM COMPLANATUM (RUD. 1814) (=C. MARGINATUM RUD. 1819).

23.

Caudal Skeleton of Bermuda Shallow Water Fishes. I. Order Isospondyli: Elopidae, Megalopidae, Albulidae, Clupeidae, Dussumieriidae, Engraulidae.[1]

GLORIA HOLLISTER.

Department of Tropical Research.

(Text-figures 1 to 53).

OUTLINE.

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INTRODUCTION.

The following paper is a study of the caudal skeleton of the Bermuda isospondylids. It deals principally with the adult fish but when young specimens were available these have been included. The Isospondyli of Bermuda are represented by six families, eight genera, and eight species.

In the genus *Harengula* no specific name has been given to the material examined. These fish have at various times been called *H. macrophthalmus* or *H. sardina,* and their proper specific determination is a problem for future study.

This study was made from specimens which were cleared by potassium hydroxide and stained by alizarin. Alizarin is a vital stain for bone and the determination of the presence and position of bones is greatly facilitated by its use. The term KOH, which stands for potassium hydroxide, the clearing chemical, was adopted in our field laboratory at Nonsuch, Ber-

[1] Contribution No. 515, Department of Tropical Research, New York Zoological Society. Contribution from the Bermuda Biological Station for Research, Inc.

muda, as a designation for specimens cleared and stained. They are numbered as such in a KOH catalogue. This term appears in the list of catalogue numbers and in general usage. For details of this clearing process see "Clearing and Dyeing Fish for Bone Study," *Zoologica*, Vol. XII, No. 10, and "Fish Magic," in the *Bulletin* of the New York Zoological Society for March-April, 1930. (Vol. XXXIII, No. 2).

The length of specimens in this paper is standard length, unless otherwise stated.

I am especially indebted to Mr. and Mrs. George Arents, Jr., and Mr. and Mrs. Bernard Baruch, Jr., for specimens of large *Tarpon*, and to the American Museum of Natural History for a specimen of *Elops*. I thank for their cooperation Dr. William Beebe and Mr. John Tee-Van of this department. The drawings are by Edward Delano, George Swanson, and Helen Tee-Van.

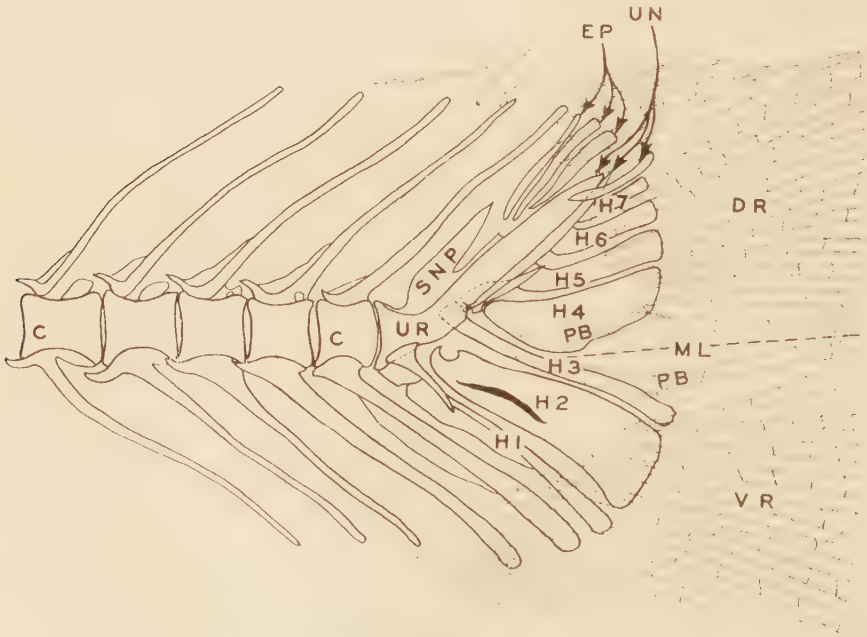
Caudal Fin Terminology.

(Text-figs. 1, 2).

Caudal Ray Count: The dorsal and ventral counts of fin rays are considered separately. The dorsal count begins anteriorly and continues around the fin to the median division in the rays. The ventral count begins anteriorly and continues around the fin to the median division. The count is expressed as follows:

$$\frac{\text{Dorsal raylets+rays=Total:}}{\text{Ventral raylets+rays=Total:}} \quad \text{or} \quad \frac{2+10}{2+10} \quad \frac{12}{12}$$

Caudal Ray: A branched (usually) caudal element possessing one or more transverse joints.

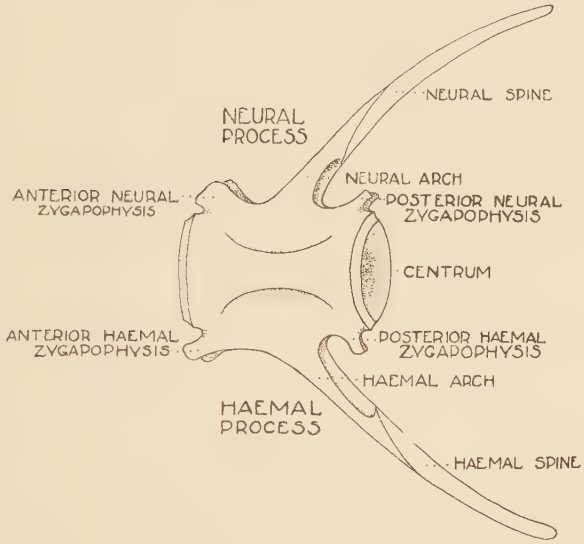


Text-figure 1.

Typical tail of a Bermuda Isospondyli. C, centrum; DR, dorsal ray; EP, epural; H, hypural; ML, median line; PB, prolonged base; SNP, specialized neural process; UN, uroneural; UR, urostyle; VR, ventral ray.

Caudal Raylet or *Simple Ray:* An unbranched caudal element possessing no transverse joints. Raylets are always anterior to the rays.

Caudal Region: The vertebral column is divided into a trunk and caudal region. The caudal region begins where the first or anterior haemal process forms a closed haemal arch with a single haemal spine projecting. Ribs are absent in the caudal region.



Text-figure 2.
Typical caudal vertebra.

Centrum: The central element of a vertebra on which the neural and haemal processes are formed.

Epural: Any bone, or bones, that are dorsal to the urostyle and support one or more caudal fin rays or spines. The bases are always unattached.

Haemal Arch: The arch on the ventral surface of a vertebral centrum through which passes the haemal vessel.

Haemal Process: The haemal arch and haemal spine on the ventral surface of a vertebral centrum.

Haemal Spine: The ventral projection below the haemal arch.

Hypural: Any bone that is ventral or posterior to the urostyle and supports one or more caudal fin rays or raylets. An expanded haemal spine. The hypural count is made from the anterior ventral part of the urostyle around to its posterior and dorsal end. The anterior hypural is called the first hypural.

Median Caudal Line: The median line is the natural median division seen in the caudal rays. This line determines whether the rays are dorsal or ventral.

Neural Arch: The arch on the dorsal surface of a vertebral centrum through which passes the nerve cord.

Neural Process: The neural arch and the neural spine on the dorsal surface of a vertebral centrum.

Neural Spine: The dorsal projection above the neural arch.

Uroneurals: The uroneurals are paired bones which are directed upward and backward on the lateral and dorsal surfaces of the urostyle. They probably represent specialized neural processes so developed to protect the sharply upturned caudal. This term is adopted from Regan (1910, 2).

Urostyle: The posterior terminal vertebral segment or segments which follow the last undoubted centrum. The urostyle is considered as one in the total vertebral count.

Zygapophysis: The dorsal and ventral articulating process of a centrum.

The inserted key illustrates the outstanding differences and similarities found in the study of the caudal skeleton of the Bermuda isospondylids. It will be noted that the three clupeids (*Harengula, Opisthonema,* and *Sardinella*) are included under one caudal pattern. Further study with more material is necessary in these species to determine the status of the three forms as shown by the caudal skeleton.

Elopidae.

Elops saurus Linnaeus.

(Text-figs. 14, 15).

Diagnostic Characters:

9 hypurals.

2 reduced posterior neural processes on the two anterior urostyle centra. Dagger-shaped.

4 distinct pairs of uroneurals. Origin of anterior pair on first or anterior urostyle centrum.

Vertebral count:

49 + 26 = 75. Haiti specimen.
57 + 24 = 81. Gravesend Bay, N. Y., specimen.
43 + 29 = 72. Jordan and Evermann, "Fishes of North America."
78-79. *Elops saurus* and *affinis*. Regan 1909.

This variation may, with further study, be correlated with geographical distribution.

Material Studied.

The following description is taken from two specimens: one caught in Haiti, Cat. No. 7172, KOH Cat. No. 2030, length 258 mm., from which this description has been made, and one caught at Gravesend Bay, which was kindly supplied by the American Museum of Natural History, Cat. No. 669, KOH Cat. No. 2136, length 280 mm. *Elops saurus* is included with the group of Bermuda Isospondyli on the basis of a single record, that of E. Linton in 1908, "Notes on Parasites of Bermuda Fishes," Proc. U. S. Nat. Mus., XXXIII, No. 1560.

Caudal Osteology.

Urostyle Centra: Four centra form the upturned caudal end of the vertebral column. The anterior three are regularly shaped. The fourth (and posterior) one is an irregular thin bony rod which curves upward posteriorly and extends as far as the anterior dorsal edge of the 7th hypural. This may represent three centra fused as it extends over the bases of three hypurals. A cartilaginous notochord extends into the dorsal contour, 1 mm. beyond the dorsalmost hypural, and is embedded between the bases of the 11th dorsal ray, above the median line (Text-fig. 15). There are about 35 horizontal striae in the cartilaginous notochord. Embedded between the striae spool-shaped centra can be seen. This condition resembles that found in the young rather than the adult *Tarpon.*

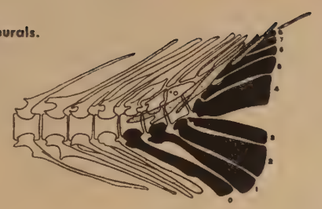
Uroneurals: This term is adopted from Regan (1910.2). There are four pairs of uroneurals, which correspond to ancestral posterior neural

Key to Caudal Fin of Bermuda Shallow Water Isospondylid Fishes. (Text-figs. 3-13).

ISOSPONDYLI

Group I

9 hypurals;
4 pair of uroneurals.



ELOPIDAE
Elops saurus

Text-figure 3.

Group II

8 hypurals;
3 pair of uroneurals.

MEGALOPIDAE
Tarpon atlanticus

Text-figure 4.

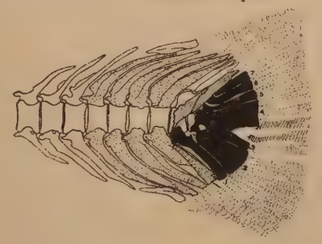
Group III

7 hypurals;

Sub-Group A

Massive neural and haemal spines on the 4 posterior vertebrae; no prolonged ray bases of the 2 median caudal rays.

2 pair of uroneurals.

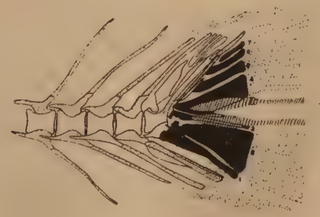


ALBULIDAE
Albula vulpes

Text-figure 5.

Sub-Group B

Slender neural and haemal spines on the 4 posterior vertebrae; prolonged ray bases of the 2 median caudal rays.



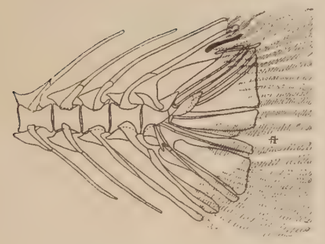
Text-figure 6.

Division I

1 epural.
Caudal rays 26, (14/12).

Text-figure 7.

DUSSUMIERIIDAE
Jenkinsia lamprotaenia

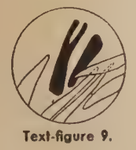


Text-figure 11.

Division II

More than 1 epural; 2, 2½, or 3.
Caudal rays 33 to 37.

Text-figure 8.



Text-figure 9.

Text-figure 10.

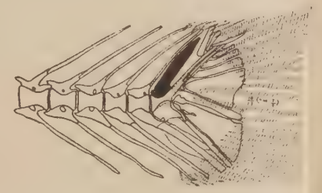
Sub-Division A

ENGRAULIDAE
Anchoviella choerostoma

Long neural process of urostyle extending dorsally at least one half the length of the anterior epural.

First closed haemal arch above anterior spine of anal fin.
Caudal and trunk vertebrae almost evenly divided.

Caudal rays 33 to 37, 17/16 to 19/18, never 19/16 as in Sub-Division B, Clupeidae.



Text-figure 12.

Sub-Division B

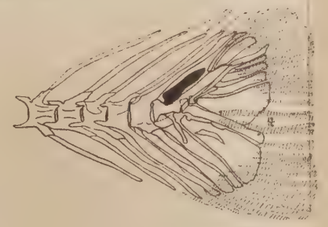
CLUPEIDAE

Short neural process of urostyle extending to the ventral tip, or slightly beyond, of the anterior epural.

First closed haemal arch well in advance of the anal fin.

More caudal vertebrae than trunk.

Caudal rays 35, 19/16.



Text-figure 13.

Group A

25 to 26 haemal arches.
(Total vertebral count 37 to 40).

Harengula sp.
(See Text-figs. 47 and 48).

Group B

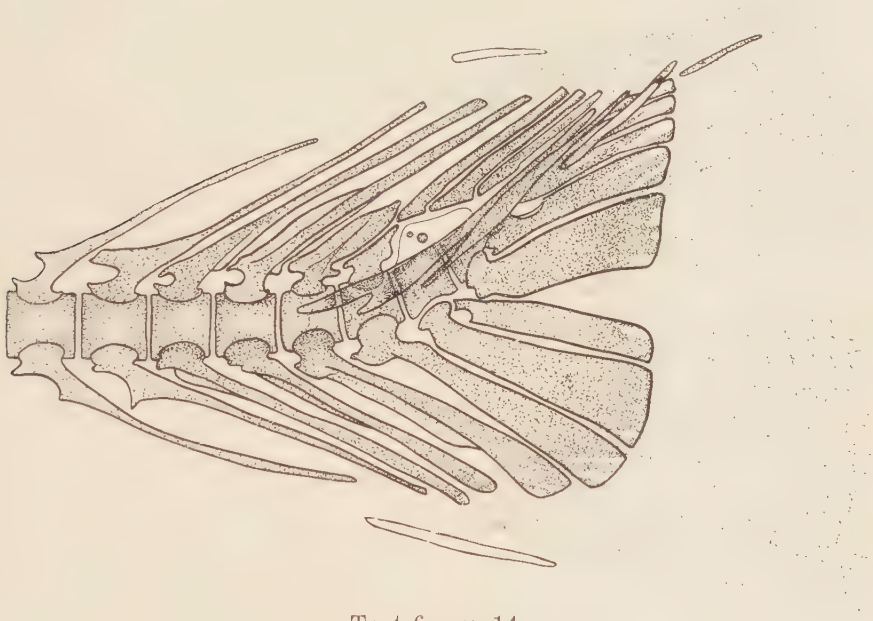
29 to 32 haemal arches.
(Total vertebral count 45 to 46).

Opisthonema oglinum
(See Text-figs. 49, 50 and 51).

Sardinella anchovia
(See Text-figs. 52 and 53).

processes. All four are elongated and crowded together on the lateral and dorsal surfaces of the upturned centra.

The uroneural whose position is the most anterior and the most dorsal is the longest (16 mm.) and the stoutest (1.5 mm. at the deepest point) and extends from the first upturned centrum as far as the center of the anterior base of the 9th (dorsalmost) hypural. Here it terminates in a blunt tip. Anteriorly this uroneural is forked, the dorsal part commencing on the first urostyle centrum which is the fourth from the last. According to Regan (1910.1) "the forking indicates the compound nature of this bone,



Text-figure 14.
Elops saurus. Tail of a 258 mm. specimen (x 2.8).

and in some specimens the line of junction between the two component elements can be clearly seen." In our Haiti specimen of 258 mm. and Gravesend Bay specimen of 280 mm., the line of junction is not evident. The anterior pair of uroneurals is the only one that covers in part the dorsal, as well as the lateral surfaces of the urostyle. The anterior half is entirely lateral but above the posterior, or last, centrum the lateral parts meet dorsally, but do not fuse, and extend for a short distance covering both the dorsal and upper lateral surfaces of the urostyle.

The bones of the second pair of uroneurals are spindle-shaped, 12.5 mm. long and 1 mm. deep, unforked and extend along the ventral surfaces of the first pair. The second pair arises anteriorly on the third centrum and ends between the bases of the 11th caudal ray, extending 2 mm. beyond the first pair.

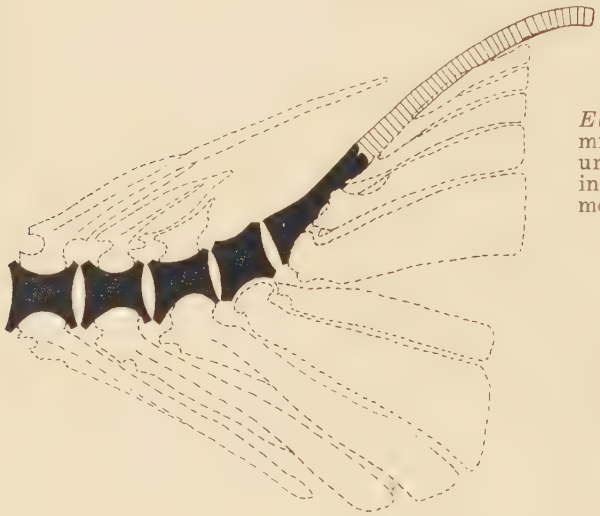
The third pair resembles the second in shape but the bones are one-half the size. The length is 6.43 mm. and the depth .43 mm. This pair arises at the posterior third of the second pair and extends slightly beyond it, ending posteriorly between the bases of the 11th ray.

The fourth pair is the smallest, length 3.86 mm. and depth .29 mm. The bones extend obliquely across the 10th dorsal ray with the anterior tips

on the dorsal side and the posterior on the ventral side of the ray. The direction does not parallel that of the other three pairs; instead there is a decided posterior slant. The entire length of the two bones is free in the tissue that covers the ray bases; whereas the dorsal tips of the other three uroneurals are between the bases of the 11th caudal ray.

None of these pairs are fused one with the other. The two bones of each pair are distinctly individual.

Hypurals: There are nine hypurals on the urostyle, four below and five above the median line. Two additional anterior long haemal spines project into the ventral caudal contour with fin rays attached. The third hypural (counting from the anterior to the posterior dorsal) is the largest.



Text-figure 15.
Elops saurus. Tail of 258 mm. specimen with paired uroneurals removed showing, in black, the vertebral segments of the urostyle (x 3).

The ninth and dorsalmost hypural is the smallest. The bases of the third and fourth hypurals are ventral and adjacent to a single centrum. This is also shown in Regan's drawing (1910.1) and is present in *Tarpon* and *Albula* and indicated in all the Bermuda isospondylids. None of the hypurals are fused. All of the bases are cup-shaped and the two dorsal ones are more pronounced, almost bifid, the basal tips extending on either side of the notochord. The Haiti specimen is more developed in this respect, the bases being noticeably longer. (Text-fig. 17 for example in *Tarpon*).

Epurals: There are three epurals, all long flat bones expanded at the ventral ends. The first and anterior epural is the longest, its ventral end slightly over-lapping the tip of the anterior reduced neural process.

Specialized Neural Processes: There are two reduced neural processes on the first and second upturned urostyle centra. Both are short, broad, dagger-shaped bones, the anterior one being the larger. The anterior neural process is on the first centrum that shows a tendency to turn upward. So this centrum is considered the first of the urostyle series and the attached haemal spine is called the first hypural in this paper.

The shape of the neural processes of the Gravesend Bay specimen is different from the Haiti specimen, the tips being blunt and the same depth as the rest of the process. There is a third smaller process, which is not present in the Haiti specimen. This may be an individual irregularity and not a common variation. But as there are only two specimens from which to draw conclusions this point will be left open to be determined later. This study is too new to base specific differences on characters such as the

neural processes without more specimens to establish the normal range of variation.

Dorsal to the third urostyle centrum are two small round islands of bone surrounded by a median plate of cartilage. In Regan's illustration a solid bone is shown filling this entire area below the three epurals. The Gravesend Bay specimen has one round island of bone which is of interest because of the smaller size of the fish. Regan's specimen is undoubtedly older than both of ours, although no length is given. In certain fishes of other families where a series of specimens is available for study, ranging in development from young to adult, this particular area is one of the last to ossify. Our adult six-foot *Tarpon* is a striking example.

Caudal Fin Ray Count:

258 mm. $\frac{2 + 16 = 18}{1 + 15 = 16}$ Haiti specimen. There are two dorsal and one ventral anterior raylets which lack the characteristic cross bars of the rays.

280 mm. $\frac{2 + 17 = 19}{1 + 15 = 16}$ Gravesend Bay specimen.

Specialized Ray-scales: Partly covering the first dorsal and ventral raylets there is a thick elongate bony ray-scale. This was present in *Leptolepidae,* and of the Bermuda isospondylids, is seen in a more reduced size in *Tarpon, Albula,* and most of the clupeids.

MEGALOPIDAE.

Tarpon atlanticus (Cuvier & Valenciennes).

(Text-figs. 16-20).

Diagnostic Characters:

8 hypurals.

Small pointed reduced neural process on the anterior part of the first urostyle centrum.

3 distinct pairs of uroneurals.

Vertebral count: 33 + 24 = 57.

In the closely related Pacific and Indian Ocean *Megalops cyprinoides,* the vertebral count is 38 + 30 = 68. (Delsman, 1926).

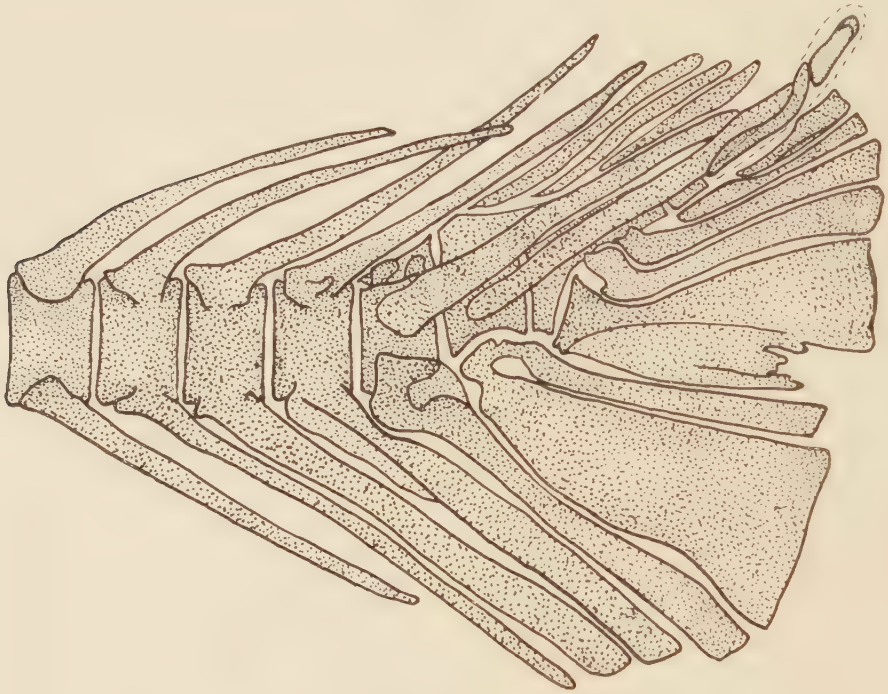
Material Studied.

The following description is from one adult fish, weight one hundred pounds, length six feet, or 1,800 mm. (Text-figs. 16, 17, 18). This specimen was taken in Florida, the gift of Mr. and Mrs. George Arents, Jr., KOH Cat. No. 2085. Any differences in younger stages are mentioned, being described from a two-foot fish, 635 mm., caught in Florida, the gift of Mr. and Mrs. Bernard Baruch, Jr., KOH Cat. No. 2083, and from three specimens, (Text-figs. 19, 20), taken in Haiti, Cat. No. 7303, KOH Cat. Nos. 2031 and 2033, lengths 140, 120, 115 mm. At the time of writing (October, 1936), the 115 mm. specimen is the smallest *Tarpon* available for study in the collection of the Department of Tropical Research and in all other institutions with which I communicated. The two-foot specimen is essentially like the six-foot fish and the drawing for the latter represents both stages and all those in between.

In Bermuda *Tarpon* are rare. A single skin was seen by G. Brown Goode, (Catalogue of the Fishes of the Bermudas, 1876), in the collection of John T. Bartram of St. George. We have seen *Tarpon* only occasionally while helmet-diving.

Caudal Osteology.

Urostyle Centra: In the six-foot fish two complete centra, one elongate centrum and an additional reduced terminal centrum, form the urostyle. The elongate element extends from the fourth hypural to the tip of the seventh. This is heavier and larger than in the younger specimens. It is



Text-figure 16.
Tarpon atlanticus. Tail of 1,800 mm. specimen (x 4/5).

irregularly shaped throughout and in the center of its length on the ventral side it resembles a thin keel which lies between the pseudoarches of the fifth and sixth hypurals. The terminal bony segment, which is not present in the smaller specimens, extends three-fourths of the length of the base of the seventh hypural. It is open above but complete below and the ossification is thin and delicate (Text-fig. 16). The notochord is seen extending from this last ossified segment into the caudal contour 10 mm. beyond the dorsal, or eighth hypural. Its end is embedded in the eleventh dorsal ray above the median line. Enclosed in a tough fibrous sheath are 18 or more separate irregular vertebral elements. All are slightly ossified. The ossification of the tip end which extends beyond the hypurals is heavier on the edges and the tip than in the center. Ventrally, this seems to be solid but dorsally it appears cleft and may be two lateral plates in close proximity (Text-fig. 18).

In the smallest specimens (Text-fig. 19), two complete centra and one elongate terminal centrum form the urostyle. This posterior rod-like centrum shows definite indication of fusion of two centra. As it extends over three hypurals, the fourth, fifth, and sixth, similar to *Elops*, it may be a composite of three centra. The cartilaginous notochord prolongation from this last centrum, very similar to that of *Elops*, extends from the base of

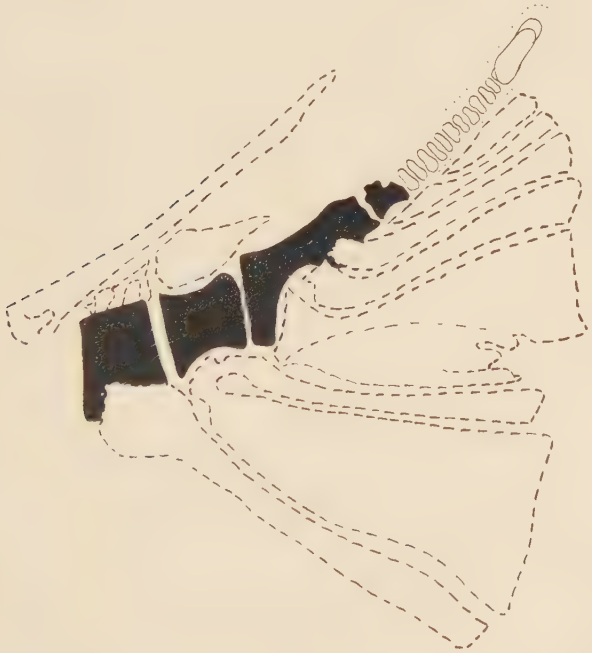
the sixth hypural into the dorsal caudal contour, 3 mm. beyond the eighth hypural (Text-fig. 20). As in *Elops*, it is embedded in the base of the 11th dorsal ray, above the median line.

Uroneurals: There are three pairs of uroneurals, which correspond to ancestral posterior neural processes. All three are elongated bones and are close together on the dorsal and lateral surfaces of the upturned uro-

Text-figure 17.
Tarpon atlanticus. The eighth hypural showing the bifid, arch-like base in three-quarter view (x 1).

style, covering the three centra and the cartilaginous notochord, all except the extreme tip which extends beyond the hypurals. None of these pairs is fused one with the other and the two elements of each pair remain distinct.

The first uroneural, anterior and dorsalmost in position, is the longest (67 mm.) and the widest (8 mm.) at the deepest part, which is the anterior end. (The measurements of the small specimens are 11 mm. by .8 mm. wide). The bones of this pair extend from the anterior edge of the first centrum of the urostyle, above, but on a vertical line with the center of the dorsal side of the 8th hypural. They end in tapering pointed tips. The an-



Text-figure 18.
Tarpon atlanticus. Tail of 1,800 mm. specimen with paired uroneurals removed showing, in black, the ossified vertebral segments (x 4/5).

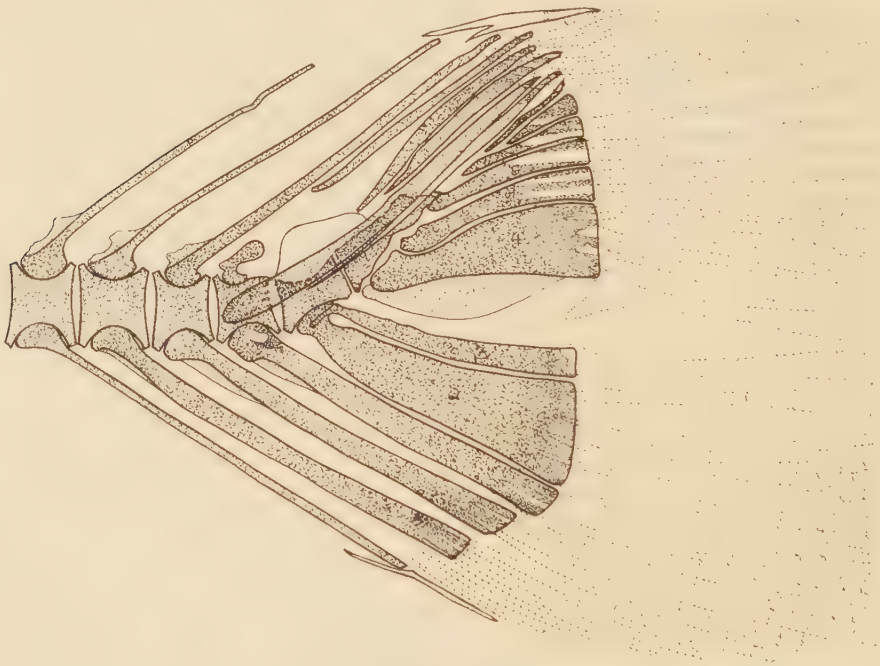
terior is rounded and unforked and differs in this respect from *Elops*. This is the only pair to cover in part the dorsal as well as the lateral surfaces of the urostyle. The anterior half is entirely lateral and above the third urostyle centrum the two lateral bones meet dorsally, but do not fuse.

The bones of the second pair of uroneurals are spindle-shaped with rounded anterior ends. This differs from the three small specimens of 115,

120, and 140 mm. as the illustration shows (Text-figs. 16, 19). The length of the uroneurals is 61 mm. and the width at the widest part in the center, is 6 mm. They arise one-fourth of the distance from the anterior edge of the second centrum of the urostyle and extend ventral and parallel to the first pair. Distally they end between the bases of the 10th and 11th caudal rays, above the median line, and extend 8 mm. beyond the first pair. (The measurements of the three small specimens are 9.2 mm. by .4 mm. wide).

The third pair of uroneurals is very small, the left bone being longer and more slender than the right and less closely associated with the underlying uroneurals. The length of the left bone is 25 mm. and the width 3 mm. It arises, approximately, at the posterior third of the second pair above the tip of the 8th hypural. The bones extend across the tips of the 2nd uroneurals, bending obliquely upward and projecting across the notochord beyond the epurals. (The measurements of the small specimens are 3 mm. by .1 mm.).

Hypurals: There are eight hypurals on the urostyle, three below and five above the median line. Three additional long haemal spines project into the caudal contour with fin rays attached. As in *Elops* there are two



Text-figure 19.
Tarpon atlanticus. Tail of 150 mm. specimen (x 4.75).

hypurals ventral and adjacent to a single centrum, the second. In *Tarpon* the basal ends of the two hypurals are closely associated in the small fish and appear as one in the two-foot and six-foot specimens. In our *Elops* the basal ends are distinct. The association of two hypurals with the 2nd centrum is also present in *Albula vulpes* where in the adult they are in close proximity to each other and the centrum. In all of the Bermuda Isospondyli, other than *Elops*, *Tarpon*, and *Albula*, the larger and ventral bone of the two below the 2nd centrum is entirely free from the centrum but the smaller and dorsal bone has become united with it.

All of the hypurals remain unfused and separate for their entire length. As in many specimens of other families, there is a noticeable band of cartilage on the extremity of all of the hypurals and in the big *Tarpon* there is a trace, here and there, of ossification on the outer edge of this band. As in *Elops*, the bases of the hypurals are cup-shaped and bifid (Text-fig. 17).

Text-figure 20.
Tarpon atlanticus. Tail of 140 mm. specimen with paired uroneurals removed showing, in black, the ossified vertebral segments of the urostyle (x 5.5).

Epurals: There are three epurals which are all long and rod-like. They differ slightly in shape and length, the anterior epural being the longest and the most slender and the posterior two being the shortest and the stoutest.

In the three small specimens there is an unossified area between the ventral tips of the epurals and the urostyle centra. There is a plate of cartilage here which, in both the two-foot and six-foot specimens, is ossified. In *Elops* this area is considerable smaller, being partly filled by the overlapping of the reduced neural processes and the tips of the epurals. In *Elops* a cartilage plate is present with two small round ossified islands in the larger and one center of ossification in the smaller.

Specialized Neural Processes: In the three small fishes there is one reduced neural process which is located on the first upturned urostyle centrum. This is a rounded hook-shaped bone which curves abruptly toward the posterior. In the six-foot specimen this hook-shaped reduced neural arch is present but in addition there is a second smaller one arising on the same centrum and both are united by and appear embedded in a median bony plate filling the area above this centrum. Possibly this is an individual irregularity or malformation during growth of this particular specimen.

Caudal Fin Ray Count:

	1,800 mm. fish.	16
		13
	635 mm. fish.	17
		14
140, 120, 115 mm.	3 + 13 =	16
	1 or 2 + 13 =	14 or 15

Specialized Ray-scales: Without the study of cleared and alizarin stained specimens, the single dorsal and ventral ray-scale would be counted as caudal rays, so perfectly do their extremities form part of the series of graduated caudal raylets. But in the cleared specimens the bases are seen to be quite unlike the rays in shape, origin, and position and resemble more the structure of the ray-scales found in *Elops* and *Albula*, and some of the clupeids, than that of raylets. Regan (1910.1) made the following note: "In *Elops*, but not in *Megalops*, there is an oblong ray-scale above and below, partly covering the first upper and lower rays."

Albulidae.

Albula vulpes (Linnaeus).

(Text-figs. 21-39).

Diagnostic Characters:

7 hypurals.

No reduced neural processes as in *Elops* and *Tarpon*.

2 distinct pairs of uroneurals in the largest adult.

4 distinct pairs in the smallest adults of 47, 40 and 36 mm.

Vertebral count in large adults:

42 + 27 = 69. Bermuda specimens.

42 + 28 = 70. Jordan and Evermann, "Fishes of North America."

47 + 27 = 74. Delsman, Java Sea specimens.

Material Studied.

	Group	*Length*	*Cat. No.*	*KOH Cat. No.*	*Text-fig. No.*
10 Adults, ranging from 560 to 22 mm.	A	560 mm.		1134	21, 29
		510 mm.		1084	21, 29
		403 mm.		815	21
		206 mm.	9647	670	21, 28
	B	87 mm.	25184	768	29
		84 mm.	25183	767	25, 26
	C	47 mm.	25181	2057	22, 23, 24
		40 mm.	25182	2060	22, 23, 24
		36 mm.	25185	2081	22, 23, 24
	D	22 mm.	25208	2120	34
1 Intermediate		26 mm.	25209	2119	33
Leptocephalus (3 out of 15)		30.8 mm		2115	32
		45 mm.		2080	31
		55 mm.		147	30

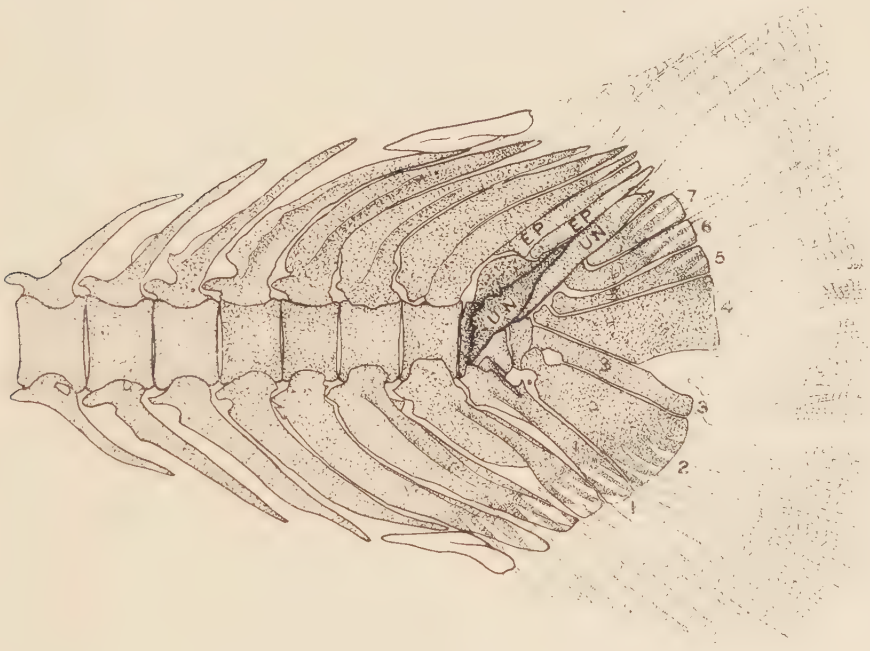
This paper dealing with isospondyls is concerned more particularly with adult specimens. *Albula* is an exception in its series of fish which range in size from young to adult. The following does not aim to be a complete description of the leptocephalus phase but rather a summary of the outstanding changes occurring in the different growth stages. During seven seasons of work in Bermuda specimens of the above range in lengths were collected and studied. The figure at the top of the column represents the largest and oldest and the last figure, 55 mm., the youngest. It will be seen by reading the column from the bottom to the top that the young in growing first decrease in length; at the same time they change from ribbon-like creatures to the shape of the adult. Then growth continues by lengthening.

Although specimens have been had in collections which show *Albula*

in both phases, it has not been known just when the change occurs and the rapidity of this change. I definitely established these facts by observations on a living *Albula* which grew and shrank from 55 mm. to 20 mm. in ten days' time. During this time it changed from the leptocephalus to the adult in body shape.[2]

Caudal Osteology.

Urostyle Centra: The fully adult urostyle (560, 510, 403 and 206 mm.) is composed of two complete centra, the posterior one being greatly reduced in size (Text-figs. 21, 29). Almost completely hidden under the uroneurals is a posterior terminal bony mass which probably represents several fused centra (Text-fig. 28). In the three small specimens of 47, 40, and 36 mm., four centra can be distinguished (Text-figs. 22, 23, 24). The two posterior ones are rod-shaped and very close together. The posterior terminal bone



Text-figure 21.
Albula vulpes. Tail of 560 mm. specimen (x 1.4).

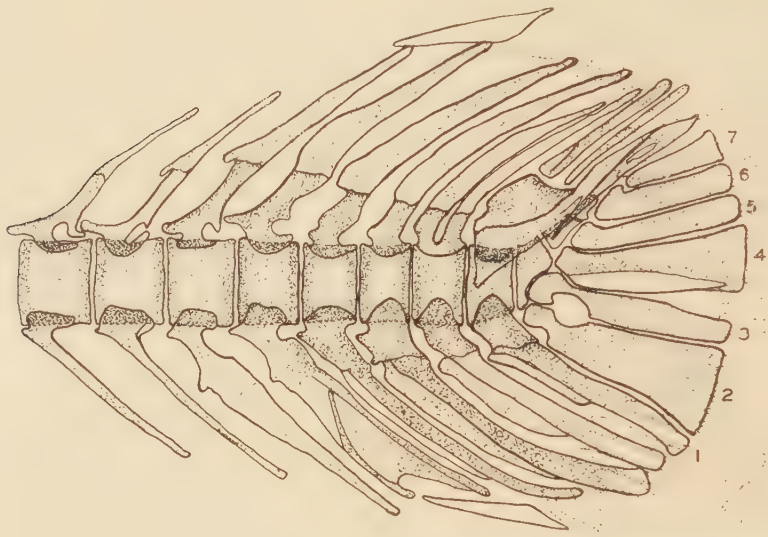
is above the center of the base of the sixth hypural (third above the median line). In the 87 and 84 mm. specimens there are only three centra, the posterior two having fused (Text-figs. 25, 26, 27).

In dissecting the 206 mm. specimen (Text-fig. 28) the second urostyle centrum was found noticeably reduced and the fourth hypural elongated with its base in the position of the third centrum as seen in the 84 mm. specimen (Text-figs. 25, 26). The terminal fused centrum is pushed out of its youthful position, where it formed the upward curve in the urostyle, and its anterior basal end is almost superimposed on the second urostyle centrum. The notochord is seen extending posteriorly as in smaller speci-

[2] See *Bulletin*, New York Zoological Society, May-June 1936, Vol. XXXIX, No. 3.

mens. Anterior to the base is a minute reduced bony element which looks like a miniature arch base. This is median in position, lying between the sides of the specialized neural process of the urostyle.

In the two largest specimens (560 and 510 mm.), no cartilaginous notochordal prolongation can be found after dissecting away the heavy uroneurals. But in all others, including the 206 mm. specimen, a delicate



Text-figure 22.
Albula vulpes. Tail representing 47, 40 and 36 mm. specimens (x 15.5).

notochord extends, within the uroneurals, into the caudal contour, ending between the bases of the eleventh caudal ray above the median line. The notochord extends to below the tip of the sixth dorsal raylet, counting from the anterior (Text-fig. 28).

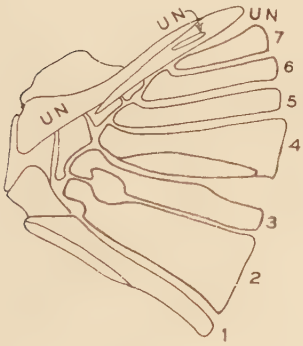
Uroneurals: In the four largest specimens (560, 510, 403, and 206 mm.) there are two pairs of heavy uroneurals which overlap each other (Text-figs. 21, 29). The irregular forward edge of the anterior pair can be traced very plainly, as it covers the greater part of the first urostyle centrum. Ventrally, its long slender tip meets the dorsal edge of the corresponding hypural. This anterior pair extends dorsally and posteriorly to about mid-length of the seventh hypural, which is the dorsalmost of the series. The posterior half is completely covered by the second pair of uroneurals, whose origin is in the center of the second urostyle centrum, dorsal to the second and third hypurals. This pair extends almost to the extremity of the seventh hypural.

In the smallest specimens (47, 40, and 36 mm.), there are four pairs of uroneurals (Text-figs. 23, 24). The two pairs which correspond to those of the adult elongate anteriorly with growth (Text-figs. 23, 29). In comparison with the 45 mm. leptocephalids (Text-figs. 36, 37), these bones have almost doubled in length, and overlap each other. In the leptocephalids only the distal and proximal ends meet. The uroneurals appear stained in the 30.8 mm. leptocephalus. In 84, 47, and 40 mm. specimens the anterior tips of the first uroneural is on the third urostyle centrum (Text-figs. 23, 25).

In the 87 mm. and all larger specimens it is on the second urostyle centrum (Text-figs. 27, 29). There is a third pair seen only in leptocephalus stages and 87, 84, 47, 40, 36 and 22 mm. specimens. In the leptocephalus this pair is the most dorsal and posterior (Text-fig. 36). These uroneurals do not elongate with growth and remain identical in length from the leptocephalus to the 87 mm. stage and become fused in the center of the first uroneural. In specimens of 206 mm. and over, there is no trace of this third pair (Text-fig. 29). A fourth pair is seen in the 47, 40, and 36 mm. specimens, extending in a dorsal direction posteriorly from the reduced neural process. This is not to be found in the largest or fully adult specimen (Text-fig. 24).

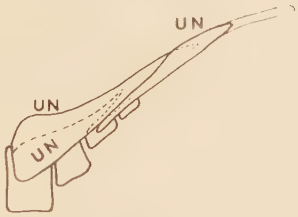
Hypurals: There are seven hypurals ventral and posterior to the urostyle, three below and four above the median line. Four additional haemal spines project into the caudal contour. The anterior one has above it the specialized ray-scale, and the three others have fin-rays attached (Text-fig. 21).

As in *Elops* and *Tarpon* the two hypurals immediately ventral to the median line arise from what appears to be a single centrum. In all other Bermuda adult Isospondyli the larger hypural of the two is well separated from any centrum attachment and is one of the largest hypurals. It resembles an isolated triangular island of bone. In the very young stages of several of the clupeids there is to be seen this same attachment to a



Text-figure 23.

Albula vulpes. Tail of 47, 40 and 36 mm. specimens showing the position of the three pairs of uroneurals in relation to the urostyle segments and hypurals (x 20).

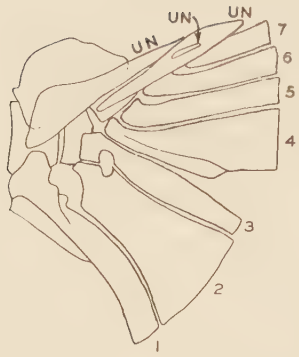


Text-figure 24.

Albula vulpes. Dissection of Text-fig. 23 showing four urostyle segments and a fourth pair of uroneurals which are dotted and lettered, being under the anterior uroneurals. The smallest pair of uroneurals has been omitted in order to show the extent of the underlying tips of the anterior uroneurals (x 20).

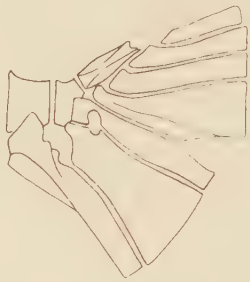
single centrum as in *Elops*, *Tarpon*, and *Albula* (Text-figs. 14, 16, 21). The bases of the hypurals are bifid as in *Elops* and *Tarpon* (Text-fig. 17, *Tarpon*).

Epurals: In the adult there are two epurals which are heavy irregular bones and in such close proximity that their respective outlines are difficult to trace (Text-fig. 21). In the young of 47, 40, and 36 mm. the two epurals are distinct (Text-fig. 22). The area above the urostyle in the young and adult is almost completely filled with the uroneurals and the specialized neural process. It is interesting to note here that in the longest and consequently the youngest of the leptocephali, where there is no evidence of



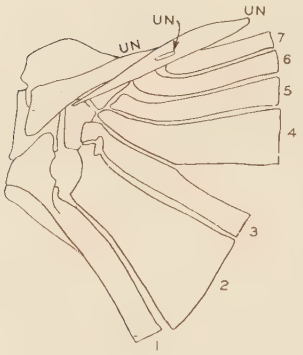
Text-figure 25.

Albula vulpes. Tail of 84 mm. specimen showing the positions of the three pairs of uroneurals in relation to the urostyle segments and hypurals. The large pairs have lengthened with growth but the small pair has remained the same (x 9).



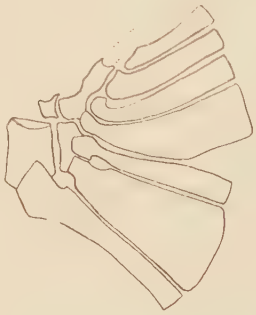
Text-figure 26.

Albula vulpes. Dissected tail of 84 mm. specimen showing three urostyle segments instead of four as seen in the younger stages (x 7.8).



Text-figure 27.

Albula vulpes. Tail of 87 mm. specimens showing the positions of the three pairs of uroneurals in relation to the urostyle segments and hypurals. The bones of the dorsalmost uroneural have lengthened and the anterior tips are on the second urostyle segment instead of the third as in smaller specimens. The length of the small pair has remained unchanged (x 8).



Text-figure 28.

Albula vulpes. Dissected tail of 206 mm. specimen showing the more consolidated urostyle and remains of the fourth uroneurals of Text-fig. 24 (x 2.5).

segmentation in the notochord nor absorption of alizarin, the basal ends of the two epurals are united (Text-fig. 36).

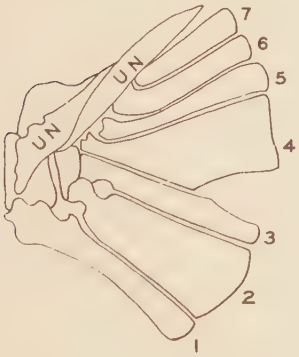
Specialized Neural Process: There is one reduced neural process on the first centrum of the urostyle (Text-fig. 21). This is ossified and de-

veloped in the 36 mm. young-adults but cannot be detected, even in cartilage form, in any of the younger specimens (Text-figs. 22, 36).

Caudal Fin Ray Count:

55 mm.	$\frac{11}{11}$	Leptocephalus. Caudal rays unstained (Text-fig. 36).
45 mm.	$\frac{11}{11}$	
30 mm.	$\frac{2 + 10 = 12}{2 + 10 = 12}$	Caudal rays stained.
26 mm.	$\frac{3 + 10 = 13}{2 + 10 = 12}$	
22 mm.	$\frac{4 + 10 = 14}{4 + 10 = 14}$	No trace of dorsal or ventral caudal ray-scale or body scales. (Text-fig. 37).
36 mm. 40 mm. 47 mm.	$\frac{6 + 12 = 18}{3 + 13 = 16}$	Caudal ray-scale present. Body scales present. (Text-fig. 22).
84 mm.	$\frac{3 + 15 = 18}{2 + 14 = 16}$	
87 mm.	$\frac{3 + 15 = 18}{1 + 15 = 16}$	
206 mm. 403 mm. 510 mm.	$\frac{1 + 17 = 18}{1 + 15 = 16}$	(Text-fig. 21).
560 mm.	$\frac{18}{16}$	(Text-fig. 21).

Specialized Ray-scales: As in *Elops* and *Tarpon,* a thick, elongate, bony ray-scale partly covers the first dorsal and ventral anterior raylet. In *Albula* this structure is heavier than in any of the other isospondylids (Text-figs. 21, 22). It is well developed in all specimens 36 mm. and longer



Text-figure 29.

Albula vulpes. Tail of largest adult showing uroneurals and segments of the urostyle reduced to two elements respectively (x 1.5).

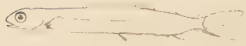
but not evident, even in cartilage form, in any of the younger stages or leptocephalids.

Additional Characters Worthy of Note: All of the posterior neural and haemal spines are very heavy and thick and by this character alone adult *Albula* can be identified and distinguished from all other Bermuda isospondylids (Text-fig. 21). It is interesting to note that in several specimens there is a double neural spine structure (Text-figs. 21, 22). This

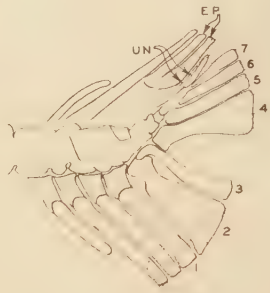
Text-figure 30.

Text-figure 31.

Text-figure 32.

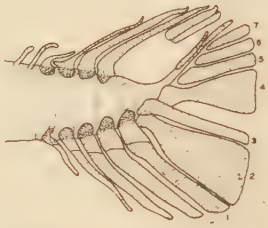


Text-figure 33.

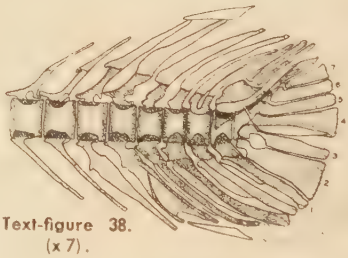


Text-figure 36.
(x 19).

Text-figure 34.

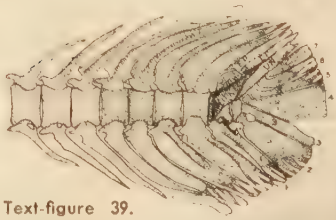


Text-figure 37.
(x 14.5).



Text-figure 38.
(x 7).

Text-figure 35.



Text-figure 39.
(x 3/5).

Text-figures 30-39.

Albula vulpes. Summary of the caudal skeleton development and corresponding change in body form from a 55 mm. leptocephalus to a 560 mm. adult. The five upper figures in the left column are natural size. Text-figure 35 is x 1/9.

occurs in specimens of the following lengths: 510, 403, 87, 47, and 36 mm. In two of these the double structure is on the next to the last, and in the other specimens it is on the last vertebra. This is not correlated with size, for in the other specimens of the series which are 560, 84, 40, and 22 mm., no double neural structure is present.

The comparative development of the skeleton is interesting in the series at hand. In the longest leptocephalus (55 mm.) the notochord is not yet segmented, nor has it taken up any alizarin (Text-fig. 36). In the successively shorter leptocephali more and more vertical lines appear at regular intervals which are the antecedents of the adult centra (Text-fig. 37). In all of the small specimens having the form of the adult, 22 mm. and larger, there is vertebral differentiation which is less distinct in the smallest specimen. Here the centra have not yet developed the shape of the adult centra. The vertebrae in the smallest specimens are rectangular and about twice as deep as wide (Text-figs. 21, 22). In all the small specimens there is ossification in the head and caudal regions. In the smallest fish of 22 mm. the only ossification of the notochord is in the caudal region, on the dorsal and ventral surfaces, near the bases of the neural and haemal spines.

It is consistent with the digging habits of *Albula* that one of the first areas of ossification should be the snout and head.

The first appearance of scales is in the 36 mm. specimen where they appear heavily stained. No scales are apparent on the 22 mm. fish.

The first appearance of ossification in the notochord is in the 22 mm. fish, and the urostyle segments are more definitely defined than in the younger intermediate 26 mm. specimen.

The dorsal fin-fold is absent for the first time in the 22 mm. specimen (Text-fig. 34).

It is interesting to note the rapidity of development which occurs between the 30.8 mm. leptocephalus and the 20 mm. young. In the *Albula*[3] that lived for ten days it took exactly six days to grow from 30 to 20 mm. It is just here during the life span of *Albula* that the unossified ribbon-like leptocephalus changes into an ossified, compact fish. In the shortening of the length the embryonic fin-folds disappear, the dorsal and anal fins and anus move forward (Text-figs. 32, 33, 34).

In the 26 mm. specimen, which is here termed an intermediate stage, change in the external shape of the body is more advanced than that of the internal and caudal skeleton, (Text-fig. 33). This resembles the 30.8 mm. leptocephalus more than the 22 mm. older form. In the 22 mm. fish the external body form resembles that of the adult (Text-figs. 34, 35). Again the development of the body is more advanced than that of the internal and caudal skeleton. In this stage is the first appearance of ossification around the notochord.

TABLE I.

Records of Extreme Lengths	*Leptocephalus*	*Adult*
From Literature	85 to 40 mm.	44 to 1220 mm.
From Bermuda Specimens	55 to 30.8 mm.	20[3] to 560 mm.
	Intermediate 26 mm.	

[3] This specimen is not cleared and stained, as it is the fish reared from a 55 mm. leptocephalus to a 20 mm. young *Albula*, where the body resembled the adult. It will be seen from the figures stated above in literature that this specimen probably is the smallest *Albula vulpes* in any collection. See the New York Zoological Society *Bulletin*, May-June 1936, Vol. XXXIX, No. 3.

DUSSUMIERIIDAE.

Jenkinsia lamprotaenia (Gosse).

(Text-figs. 40-44).

Diagnostic Characters:

7 hypurals.

1 reduced neural process. This is situated on the anterior part of the urostyle and, unlike *Elops* and *Tarpon*, it is forwardly directed.

3 distinct pairs of uroneurals.

Vertebral count: 27 + 16 = 43.

1 epural, the only isospondylid with one.

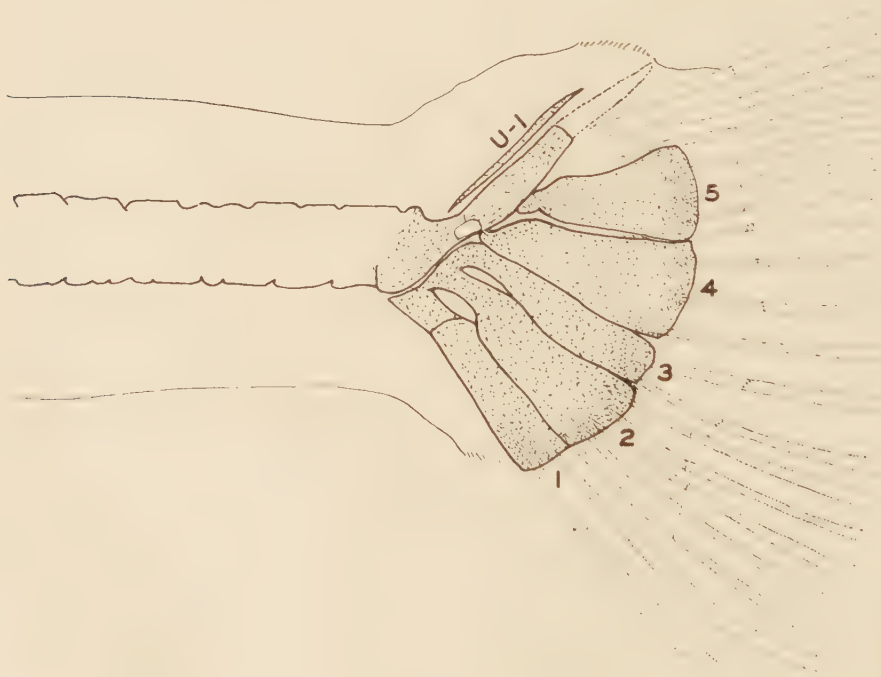
Prolonged bases of the two median caudal rays.

Total caudal ray count of 26, the smallest of the whole group.

Specialized neural processes unlike those of all other Bermuda isospondylids.

Material Studied.

This description is taken from the five following specimens and com-



Text-figure 40.

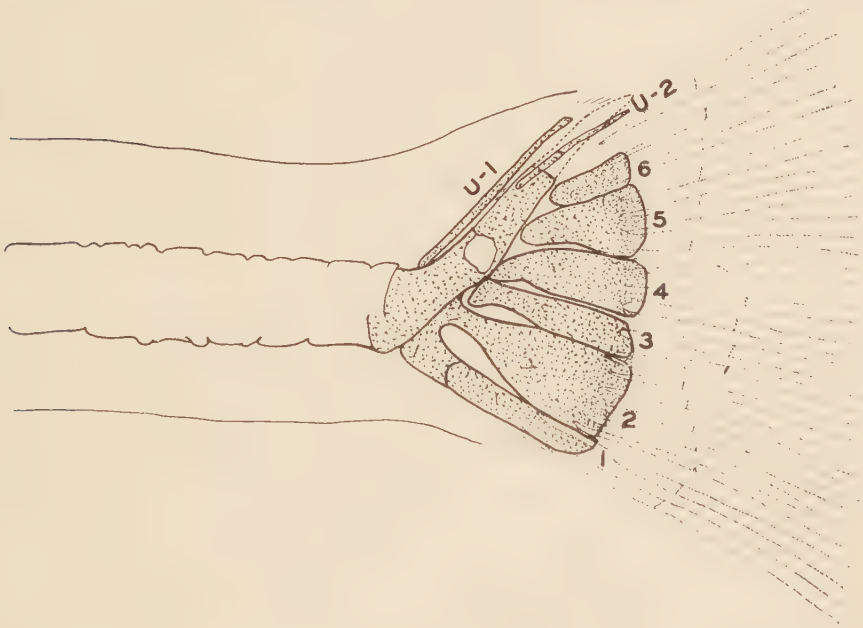
Jenkinsia lamprotaenia. Tail of 10 mm. specimen with unossified and unsegmented vertebral column. There are only five hypurals and one pair of uroneurals ossified (x 140).

parative studies were made with fifty-five additional fish which range in size from young to adult.

Length	*KOH Cat. No.*	*Text-fig. No.*
40 mm.	2096	
26 mm.	343	44
18 mm.	657	43
15 mm.	657	42
11 mm.	657	41
10 mm.	657	40

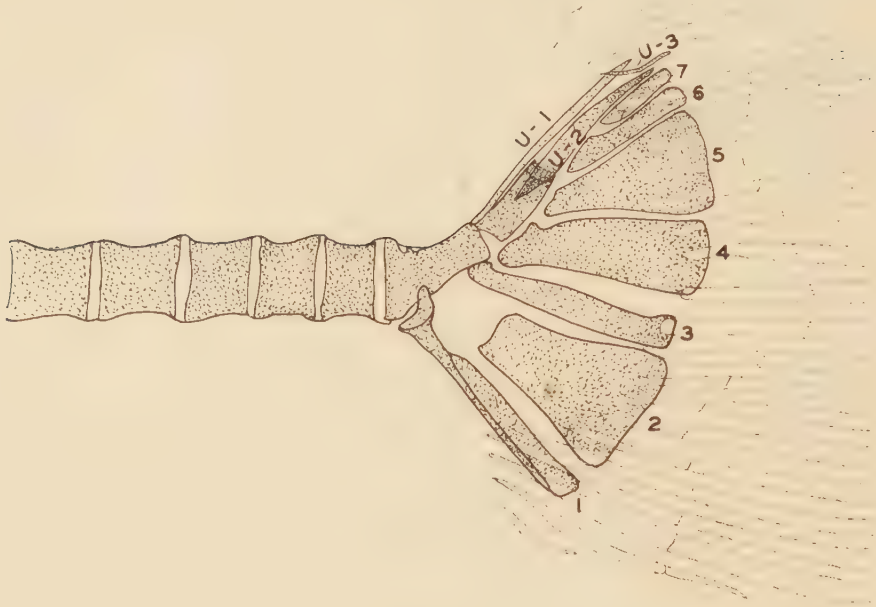
Caudal Osteology.

Urostyle Centra: The adult urostyle appears as one bone. Anteriorly it is shaped like a half centrum and posteriorly it has a slender upturned end which is situated between the fourth and fifth hypurals, counting from the anterior. The distal end has projecting ventrally a fan-shaped bone that extends over the bases of the three dorsal hypurals, and covers an unossified area between the hypural bases and the end of the urostyle. In the young stages this bone is not present. It is first seen in an 18 mm. fish (Text-fig. 43). This space does not exist before the uroneurals appear and the urostyle has become fully ossified and reduced in size. In this stage the distal end of the urostyle and the bases of the hypurals almost meet (Text-fig. 43). With growth more and more space in this area appears. The urostyle is definitely divided into two parts in the young stages. The line of junction shows plainly in the adult (Text-fig. 44).



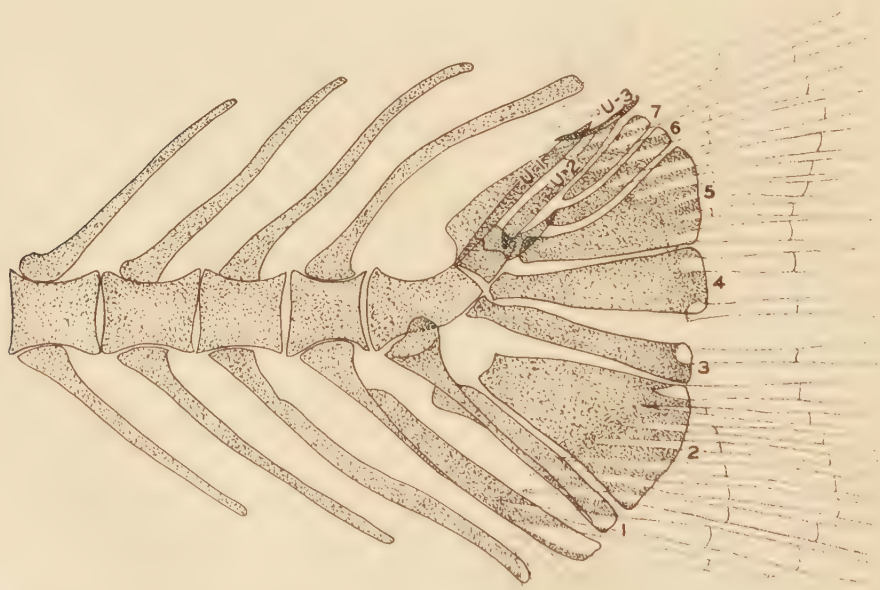
Text-figure 41.

Jenkinsia lamprotaenia. Tail of 11 mm. specimen with unossified and unsegmented vertebral column. There are six hypurals and two pairs of uroneurals ossified (x 108).



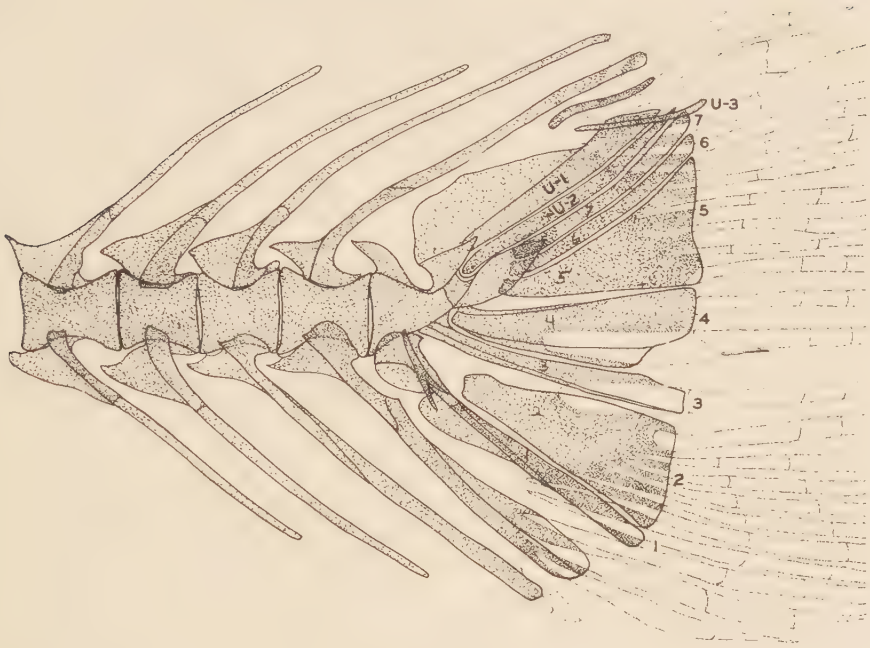
Text-figure 42.

Jenkinsia lamprotaenia. Tail of 15 mm. specimen with ossified and segmented vertebral column. There are seven hypurals and three pairs of uroneurals ossified. The basal end of the second hypural is reduced (x 64).



Text-figure 43.

Jenkinsia lamprotaenia. Tail of 18 mm. specimen with ossified neural and haemal processes (x 59).



Text-figure 44.
Jenkinsia lamprotaenia. Tail of 26 mm. specimen with single epural ossified (x 39).

Uroneurals: There are three pairs of uroneurals in the adult. Each pair is quite different in shape, size, and general position, as may be seen in the accompanying drawings. The bones are numbered according to the order of their appearance. In several young stages the first pair appear as two separate bones which in the older stages fuse into a solid structure which is directly dorsal to the urostyle. It extends from the tip of the reduced adult urostyle into the caudal contour. In the older fish of 18 mm. and larger there is a median wing-shaped bone on the anterior three-fourths of the dorsal side. In specimens of 26 and 30 mm. this bone fills all the center of the unossified area which is between the posterior neural spine, the reduced neural process on the urostyle, and the epural.

The second pair of uroneurals first appears in the 11 mm. specimen. Here the two short lateral bones extend along the cartilaginous tip of the urostyle. In a 15 mm. specimen the bones are still separate but have enlarged in length and depth. In specimens of 18, 26, and 30 mm. the two bones are united. In each example the proximal ends overlap the tip of the ossified urostyle. The two bones of the diminutive third pair remain individual in all stages. They cannot be distinguished in a 11 mm. fish but in a 15 mm. specimen they are present. Their position is between the bases of the 10th and 11th dorsal caudal rays which are above the median line. There is a noticeable space in specimens of all lengths and here the cartilaginous prolongation of the notochard extends. The posterior ends of these bones arise beyond the caudal contour of the hypurals and extend obliquely forward and downward. (Text-figs. 41, 42).

The third pair of uroneurals is first seen in a 15 mm. specimen. This is the smallest of the three pairs, and the direction of the bones is more toward the anterior than that of the others. In the 15 mm. specimen the

position is between the dorsal tips of the first and second uroneurals with the anterior end of each bone crossing the end of the first uroneural. The posterior ends extend beyond the distal edge of the hypurals and occupy the area between the ray bases where the notochord extends. The accompanying drawings show the position and lengthening with growth (Text-figs. 42, 43, 44).

Hypurals: There are seven hypurals, three below and five above the median line. Two additional long haemal spines project into the caudal contour with fin-rays attached. In eleven young fish between the lengths of 10 and 13.44 mm., the second hypural is complete and the same length as the other hypurals. In twenty specimens between the lengths of 13.58 mm. and 40 mm., the base of the second hypural is reduced. It is entirely separate and free from the other hypurals and appears as a triangular island.

Epurals: There is only one epural in *Jenkinsia* which is first seen ossified in the 26 mm. fish. This bone can be seen in the smaller specimen but it is entirely unossified. *Jenkinsia* is the only Bermuda isospondyl having but one epural.

Specialized Neural Processes: In the completely ossified specimens of 26 mm. and larger there is a hook-shaped neural process on the posterior part of the urostyle. In the center of the urostyle this becomes very narrow and again expands posteriorly. The proximal end of the first uroneural is inserted between its tip ends. Dorsal to both is a thin median wing-like bone which is first seen in the 26 mm. specimen (Text-fig. 44).

Caudal Fin Ray Count:

26 mm.	2 + 12 = 14	(Text-fig. 44).
	1 + 11 = 12	
18 mm.	3 + 11 = 14	(Text-fig. 43).
	2 + 10 = 12	
15 mm.	2 + 11 = 13	(Text-fig. 42).
	1 + 10 = 11	
11 mm.	10	(Text-figs. 41, 40).
10 mm.	9	

Additional Characters Worthy of Note: In the fully ossified specimens of 26 mm. and larger the two median caudal rays have enlarged bases which project anteriorly half the length of the hypurals (Text-fig. 44). Among the Bermuda isospondyls this character is seen also in *Anchoviella* and the clupeids.

Enlarged wings on the anterior margin of the proximal ends of the haemal and neural spines are seen first in the 26 mm. specimen (Text-fig. 44).

In the youngest specimens vertebral ossification and individual vertebrae are first seen in 15 mm. fish. Here the urostyle, hypurals and caudal rays are well ossified. In the 10 mm. and 11 mm. specimens the vertebral column is not constricted and there is no ossification anterior to the urostyle.

According to caudal pattern and characters *Jenkinsia* seems to stand apart from the other Bermuda isospondyls. Conversely the three clupeids and *Anchoviella* are closely associated by similar patterns and characters.

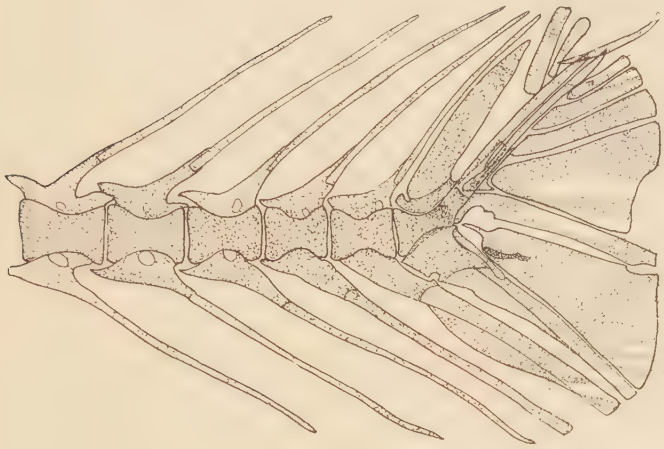
Engraulidae.

Anchoviella choerostoma (Goode).

(Text-figs. 45, 46).

Diagnostic Characters:

7 hypurals.



Text-figure 45.
Anchoviella choerostoma. Tail of 57 mm. specimen (x 11.5).

1 small reduced neural on the anterior base of the elongate neural process of the urostyle.

Vertebral count:

20 + 20 = 40. 58 mm.
21 + 20 = 41. 50 mm.
21 + 21 = 42. 42 mm.

2 epurals and occasionally 2½.

Prolonged bases of the two median caudal rays.

Material Studied.

This description is based on twenty-nine specimens ranging in size from 20 to 59 mm. The 20 mm. fish is the smallest *Anchoviella* in our collection and is identical externally with the largest (59 mm.) fish. But the internal caudal development is not the same in the young specimen, which

Text-figure 46.
Anchoviella choerostoma. Tail of 20 mm. specimen (x 28).

can be seen by the accompanying illustrations. The two specimens studied in particular are:

Length	*KOH Cat. No.*	*Text-fig. No.*
57 mm.	875	45
20 mm.	2095	46

Caudal Osteology.

Urostyle Centra: The interpretation of the adult urostyle is clarified by first studying the structure of the youngest specimen at hand. Here there are three distinct vertebral segments and the anterior part of a fourth is partly visible. (Text-fig. 46). In the large specimen, judging by the comparative position of the hypurals with that in the small fish, the two anterior segments fuse and the two posterior segments fuse. The posterior segment in the large specimen is further complicated by the presence of a small arch-like bone on the ventral side. This surrounds the proximal end of the fourth hypural. A similar structure is found in adults of the three clupeids.

Uroneurals: There are three pairs of uroneurals in the adult and the young specimen. The relative length, proximal and distal positions are the same in both fish. The anterior end of the pair that arises on the first urostyle centrum is not distinctly defined but there is little doubt that a smaller specimen would show the definite outline of the end of this bone.

Epurals: There are two and sometimes two and a half epurals in *Anchoviella.* The variation always occurs in the anterior bone which may be split in the form of a Y or have a hole in the center. This variation does not correlate with size. In fifteen specimens six have two epurals and the others have either the split or a hole in the anterior bone.

Specialized Neural Process: In the large specimens a long dagger-shaped bone extends from the anterior urostyle centrum to about the center of the anterior epural. In the 20 mm. specimen this bone is shorter and does not reach the basal end of the epural. This character, along with several others, distinguishes *Anchoviella* from the clupeids.

Caudal Fin Ray Count:

$$57 \text{ mm.} \quad \frac{5 + 13 = 18}{5 + 12 = 17}$$

In seven specimens out of fifteen, the caudal count is as stated above and is found in fish from 28 to 59 mm. In eight specimens there is variation in the total count from $\frac{17-19}{16-18}$. In the specimens examined the count of the dorsal and ventral is never $\frac{19}{18}$ as in the clupeids.

CLUPEIDAE.

1. *Harengula* sp.

(Text-figs. 47, 48).

Diagnostic Characters:

7 hypurals.

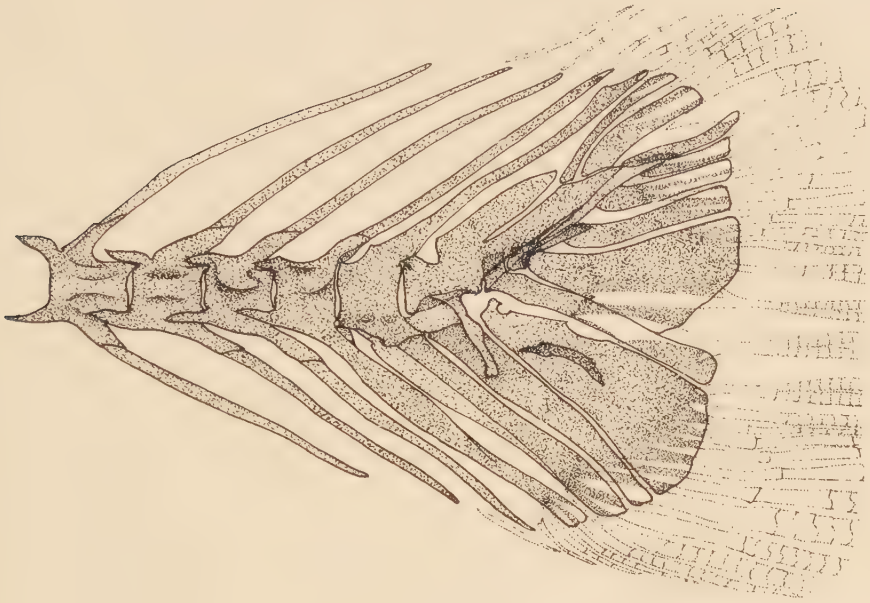
1 reduced neural process. This process is insignificant and resembles the anterior neural zygapophysis.

3 pairs of uroneurals.

Vertebral Count: 12 to 14 + 25 to 26 = 37 to 40.

2 or 3 epurals.

Prolonged bases of two median caudal rays.

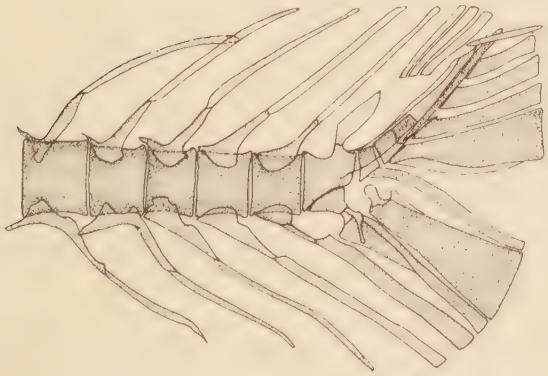


Text-figure 47.
Harengula sp. Tail of 165 mm. specimen (x 3.6).

Material Studied.

The KOH collection contains seventeen specimens ranging from 22 to 180 mm. The figure of the adult represents the tail structure of fish from 70 mm. to the largest. The illustration of the 22 mm. specimen, which is the smallest in the department collection, shows slight differences in degree of development. The two specimens studied in particular are the following:

Length	*KOH Cat. No.*	*Text-fig. No.*
165 mm.	847	47
22 mm.	656	48



Text-figure 48.
Harengula sp. Tail of 22 mm. specimen (x 21).

Caudal Osteology.

Urostyle Centra: As in *Anchoviella,* the interpretation of the adult urostyle is best made by examining the youngest specimen (Text-fig. 48). Here, as in *Anchoviella,* there are four centra or vertebral segments in the urostyle. The anterior or first and second are separate but the third and fourth appear fused. In the adult all but the anterior or first segment have become comparatively reduced. The fourth cannot be seen under the heavy uroneurals. As in *Anchoviella* the first hypural is attached to the anterior urostyle segment and the third and fourth hypurals are attached to the second and third urostyle segment.

Uroneurals: As in Anchoviella there are three pairs of uroneurals in the adult and the young specimens. The illustrations show the relative size and position of these bones.

Epurals: There are two or three epurals in *Harengula.* In seven specimens ranging from 35 to 180 mm., I found two epural bones and in five fish ranging from 22 to 173 mm. there were three epurals. The variation of a split or perforated bone in *Anchoviella* has not been found in *Harengula.*

Specialized Neural Process: In the large specimen a stout dagger-shaped bone extends from the anterior urostyle segment as far as the basal end of the anterior epural. In the 22 mm. fish this bone is shorter (Text-fig. 48). In *Anchoviella* this bone extends beyond the end of the epural.

Caudal Fin Ray Count:

165 mm.	1 + 18 = 19	(Text-fig. 47).
	1 + 15 = 16	
40 mm.	5 + 14 = 19	
36 mm.	4 + 12 = 16	

In the specimens counted the dorsal and ventral combination was constant, 19/16, and never like that in *Anchoviella.*

Additional Characters Worthy of Note: The caudal pattern of *Harengula* is very like the two other Bermuda clupeids. In the number of *Harengula* specimens examined, only minor differences have been seen, such as the variation in the number of the epurals. This variation occurs in all the clupeids and in *Anchoviella.* Most Bermuda *Harengula* key to *sardina* as described in the "Field Book of the Shore Fishes of Bermuda," Beebe and Tee-Van.

2. *Opisthonema oglinum* (Le Sueur).
(Text-figs. 49, 50, 51).

Diagnostic Characters:

7 hypurals.
Small reduced pointed neural on the anterior of the urostyle.
Vertebral Count: 16 + 29 = 45.
3 epurals.
Prolonged bases of two median caudal rays.

Material Studied.

This description is based on five specimens of the following lengths:

No. of specimens	*Length*	*KOH Cat. No.*	*Text-fig. No.*
2	110 mm.	850	
3	75 mm.	849	49, 50, 51

Text-figure 49.
Opisthonema oglinum. Tail of 75 mm. specimen (x 4.3).

These lengths represent the smallest and largest *Opisthonema* captured during our seven years in the field in Bermuda. The 75 mm. specimens are identical in development with the 110 mm. fish.

Caudal Osteology.

Urostyle Centra: This is very like *Harengula,* as the figures show. The dissection made by removing the superimposed uroneurals shows the adult elements of the urostyle. This condition is probably representative of all the Bermuda clupeids.

Uroneurals: The bones of the second pair of uroneurals are stout and noticeable heavier than those of *Sardinella* and *Harengula.* In other respects there is similarity between the three genera.

Epurals: There are three epurals found in the five specimens examined.

Text-figure 50.
Opisthonema oglinum. Tail of 75 mm. specimen dissected to show structure of the urostyle (x 3.7).

Text-figure 51.

Opisthonema oglinum. Last ossified segment of urostyle with cartilaginous notochord extending. A. View from the top; B. View from the side (x 8.5).

Specialized Neural Process: This stout neural bone is similar to that of *Sardinella* and *Harengula* in relative size and position.

Caudal Fin Ray Count: The count is identical with that of *Sardinella* and *Harengula,* 19/16.

Additional Characters Worthy of Note: In the specimens examined, *Opisthonema* differs from the other clupeids in the noticeably heavier second uroneurals and in the shape of the distal ventral surface of the fourth hypural, which is more or less even instead of having a sharp projecting point. The basal end of the second hypural is blunt instead of hammer-shaped as in *Sardinella.*

3. *Sardinella anchovia* Cuvier and Valenciennes. (Text-figs. 52, 53).

Diagnostic Characters:

7 hypurals.

Small reduced blunt neural on the anterior part of urostyle.

Vertebral Count: 14 or 15 + 31 or 32 = 46.

2½ or 3 epurals.

Prolonged bases of the median caudal rays.

Material Studied.

Seven KOH specimens have been examined, which range in size from 32 to 135 mm. The caudal pattern is identical in specimens of 50 mm. and larger.

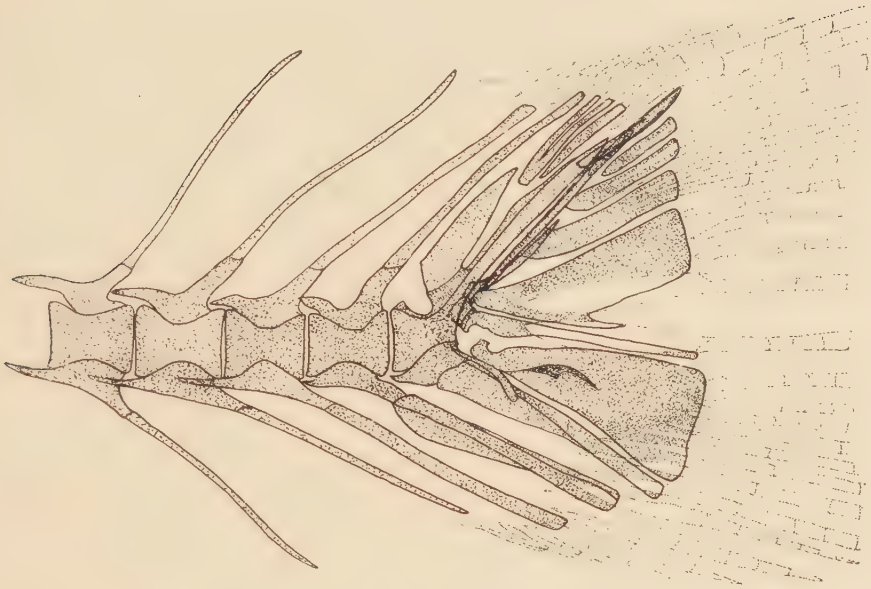
Length	*KOH. Cat. No.*	*Text-fig. No.*
60 mm. 50 mm.	671	52
34 mm. 32 mm.	2112	53

Caudal Osteology.

Urostyle Centra, Uroneurals: Almost identical with *Opisthonema* and *Harengula.*

Epurals: There are three epurals in the 32 and 34 mm. specimens. In the four larger specimens ranging from 50 to 135 mm., there are two and a half, the anterior bone being dorsally bifid. This variation is seen occasionally in *Anchoviella.*

Specialized Neural Process: This bone is almost the counterpart of that seen in *Opisthonema.*



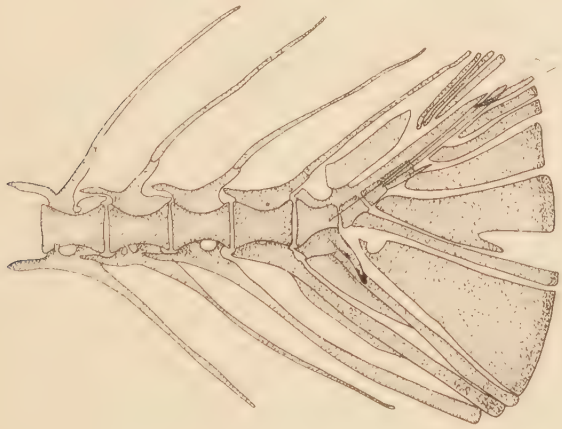
Text-figure 52.
Sardinella anchovia. Tail of 60 and 50 mm. specimens (x 28).

Caudal Fin Ray Count: As in *Opisthonema* and *Harengula,* the caudal count is 19/16.

Additional Characters Worthy of Note: The shape of the ventral distal edge of the fourth hypural differs from *Opisthonema* in that it has a sharp point. The basal end of the second hypural is hammer-shaped and this, too, differs from *Opisthonema.*

Summary.

The following paragraphs correlate the salient similarities and differences found in the study of the caudal skeletons of the Bermuda Isospondyli. According to caudal pattern and characters *Jenkinsia* seems to



Text-figure 53.
Sardinella anchovia. Tail of 34 and 32 mm. specimens (x 16.5).

stand apart from the other Bermuda Isospondyli. Conversely, the three species of clupeids and *Anchoviella* are closely associated by similar patterns and characters.

Hypurals: In the first three families described, Elopidae, Megalopidae, and Albulidae, there is no noticeable deviation of the general pattern of the hypurals from that of *Leptolepis dubius*, which is the most primitive ancestor of the Isospondyli. All the hypurals are long, expanded haemal bones. But in the adults of all the other Bermuda Isospondyli the second hypural lacks the basal part and is a triangular bone with a reduced and free base. However in the few young specimens of *Jenkinsia* available for study the primitive, ancestral, unreduced, second hypural is present and resembles that of adult *Elops*, *Tarpon*, and *Albula*.

There are nine hypurals in *Elops*, eight in *Tarpon*, and seven in *Albula*. In the first two, where the urostyle is turned up less abruptly and the segments are less consolidated than in the other Bermuda Isospondyli, there are more hypurals. It will be seen in the figures of the key that *Elops* has one more hypural than *Tarpon*, which is numbered 0 (Text-figs. 3 & 4). Also that the dorsalmost hypural which is numbered 8 in *Tarpon* is not present in *Albula*. *Albula vulpes* and the other species described in this paper have seven hypurals. Hypurals with similar numbers correspond in position in all the specimens. (Text-fig. 5).

The following table correlates the hypural count in the various genera:

	Elops	*Tarpon*	*Albula*	All other Bermuda Isospondyli
Total hypurals	9	8	7	7
Dorsal hypurals	5	5	4	4
Ventral hypurals	4	3	3	3

Epurals: All Bermuda Isospondyli have more than one epural with the exception of *Jenkinsia* which has only one. This is the last bone to become fully ossified in *Jenkinsia*.

Caudal Ray Count: With growth and increase in size the dorsal and ventral raylets change into rays, resulting in a shift of relative numbers of the two elements.

In *Albula* the total caudal count of dorsal 18 and ventral 16 remains constant in specimens ranging from 36 to 560 mm. But the dorsal and ventral raylets diminish from 6 and 3 respectively to dorsal 1 and ventral 1, with a corresponding increase of rays.

Jenkinsia has the smallest caudal count of the Bermuda Isospondyli, which is a total of 26 as opposed to 30 or more in the other species.

The long functional caudal rays are present in *Albula* leptocephali and also in very small *Jenkinsia*. With growth, additional smaller anterior rays and raylets appear.

Prolonged Median Rays: In *Elops*, *Tarpon* and *Albula* there are no prolonged bases of the two median caudal rays. All the other Bermuda Isospondyli have these bases prolonged.

Ray-scale: The ray-scale which is prominent in *Elops*, *Tarpon* and *Albula* is less obvious in the remaining Isospondyli.

General Observations: The study of a series of young *Albula* and *Jenkinsia* show that development in the caudal region commences at the posterior extremity and progresses toward the anterior. Segmentation and ossification of the notochord begins in the urostyle region and the hypurals are the first bones to appear. The neural and haemal processes increase anteriorly with growth, the first to appear being near the urostyle. In

Jenkinsia the anterior-ventral hypurals appear first and with increase in size additional dorsal hypurals appear.

In *Albula* and *Jenkinsia* the caudal skeleton becomes ossified when the fish is very small and from this stage the changes that occur during growth are changes of degree and not of kind.

In *Tarpon* and *Elops* the last part to ossify is the area directly above the urostyle. This is filled by a preformed cartilage plate. In our two *Elops* specimens of 258 and 280 mm. there are two round islands of bone within this cartilage which indicate centers of ossification. Regan's specimen has a solid bone.

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ZOOLOGICA

SCIENTIFIC CONTRIBUTIONS
OF THE
NEW YORK ZOOLOGICAL SOCIETY

VOLUME XXI

1936

Numbers 1-23

PUBLISHED BY THE SOCIETY
THE ZOOLOGICAL PARK, NEW YORK

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